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RESEARCH ARTICLE

Hepatoprotective effects of the *n*-butanol extract from *Perralderia coronopifolia* Coss. against PCP-induced toxicity in Wistar albino rats

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Abstract

In the present study, in vivo antioxidant properties of the *n*-butanol extract obtained from aerial parts of *Perralderia coronopifolia* were investigated in term of its hepatoprotective effect of female Wistar albino rats (*n*, 36; average age, 48 ± 5 days; weighing 150 ± 18 g) against PCP (pentachlorphenol)-induced toxicity. PCP (20 mg/kg b.w.) and plant extract (50 mg/kg b.w.) were administered daily by gavages for 2 weeks. Vitamin E (100 mg/kg b.w.) was given intraperitoneally as a positive control. Lipid peroxidation (LPO) levels, reduced glutathione (GSH) levels, and glutathione peroxidase (GPx) activities were evaluated in liver homogenates. While, aspartate aminotransferase (AST), alanine aminotransferase (ALT), cholesterol, and triglyceride parameters were analyzed in serums. The liver fragments were observed using light microscopy. Experimental results exhibited that PCP-treated group has a significant increase in the liver lipid peroxidation (LPO) levels of animals while decreased in plant extract-treated group. In addition, PCP caused significant decreases in glutathione peroxidase (GPx) activities and reduced glutathione (GSH) levels. Moreover, PCP induced hepatotoxicity by increasing serum transaminase enzymes, cholesterol, and triglyceride levels. While, these levels were restored to control value in animals treated with plant extract. The regularized levels of LPO, GSH, cholesterol, triglyceride, transaminase enzymes, and GPx activities revealed the antioxidant properties of the extract plant as well as of the vitamin E. The histological study showed the hepatoprotective effect of our extracts against PCP-induced acute intoxication, protecting the hepatic architecture and decreasing the functional and structural alterations of the liver. The plant extract had high antioxidant potential and completely prevented the toxic effect of PCP on the above of liver and serum parameters.

Keywords *Perralderia coronopifolia* · Pentachlorphenol · Antioxidant enzymes · Lipid peroxidation level · Glutathione · Hepatotoxicity

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Introduction

Pentachlorophenol (PCP) is an artificial organochlorine substance, prepared from different chemical components. PCP is used for the formation of fungicidal, insecticidal, and pesticide produces productions. It is also used as a wood-preserving agent at various wood treating situations (Dong et al. 2009). The oral, inhalation, and dermal interaction through a short or a long time of PCP-contact in humans and investigational animals made known distinct health effects. PCP could exert its troublemaking on respiratory, hepatic, renal, reproductive, central nervous system, endocrine functions (Wen et al. 2019; Peng et al. 2017), immune system, and thyroid homeostasis (Yu et al. 2014). Xenobiotics like pesticide have been able to yield reactive species (ROS), which in occasion cause oxidative stress in diverse tissues (Mehta et al. 2009).

However, medicinal plants have potential antioxidant properties that can help prevent the formation of free radicals and various pathologies. Phenolic and polyphenolic compounds as well as flavonoids are the most important active compounds with these properties (Sedighi et al. 2017).

Several studies have analyzed the protective effect of different medicinal plants on pesticide-induced toxicity. In this context, many medicinal plants have been tested as antioxidants in order to control the potential harmful effect of free radicals and to reduce the damage caused by pesticides. By way of example, the protective effect of the *n*-butanol extract of the *Paronychia argentea* plant (Zama et al. 2007), the methanolic extract of the *Punica granatum* plant (Agha et al. 2013), and the ethanolic extract of *Meconopsis integrifolia* (Zhou et al. 2013). Therefore, there is a growing interest in the use of natural antioxidants as a protective strategy against the liver, cardiovascular, renal, and other problems. Animal experiments have suggested the antioxidant and protective effect of some plant extracts in PCP-induced toxicity attributed to the huge amount of polyphenols. The effects of plant extracts come from their composition in bioactive molecules such as polyphenols and flavonoids (Agha et al. 2013).

Algeria contains up to 3000 species and 1000 genderswith a significant diversity of flora (Bouabdelli et al. 2012). Among them, we investigated one endemic plant; it is *Perralderia coronopifolia* Coss. from Asteraceae family. It is considered as a traditional therapeutic remedy, grows in the north-western of Africa (Boussaha et al. 2015). In vitro investigations documented that the aerial parts of this plant have a high antioxidant and anticancer ability against some cancer cell lines (Boussaha et al. 2015), as mentioned its capacity to protect DNA-damage against UV-photolysis of H_2O_2–induced oxidative damage (Bekhouche et al. 2018).

The purpose of this investigation is to evaluate the capacity of *n*-butanol extract obtained from *P. coronopifolia* and vitamin E to modulate PCP-induced liver toxicity and oxidative impairment in female rats, and whether this capacity is arising from its antioxidant activity. Therefore, we studied in vivo the antioxidant capacity of this extract via modern pharmacology testing practices. Hepatic damage was appraised by determining of serum parameters of alanine aminotransferase (ALT), aspartate aminotransferase (AST), cholesterol, and triglycerides. While the oxidative damage was estimated by measurement of malondialdehyde (MDA as oxidant agent) for lipid peroxidation (LPO) level and reduced glutathione (antioxidant agent) levels as well as glutathione peroxidation (GPx) activity. In addition, we observed the PCP-effect on the hepatic architecture using light microscopy for histological studies.

Material and methods

Chemicals

The chemicals which were used for the assays were in methodical grade and obtained from Sigma–Aldrich and Roche.

Plant material

The plant material was collected in 2011 from Taghit, Algeria. It was validated by M Mohamed Benabdelhakem, director of the nature preservation agency of Bechar. A voucher sample (PCA0511-TAG-ALG-52) has been placed at the herbarium of VARENBIOMOL Research Unit, University Frères Mentouri (Constantine 1, Algeria).

Extraction

The air-dried and ground into a coarse powder aerial parts (leaves and flowers 1400 g) of the plant were macerated for 48 h at room temperature with EtOH-H_2O (80:20, v/v), three times. After filtration, the filtrates were combined and concentrated under reduced pressure (up to 35 °C). The remaining solution (500 mL) was dissolved in H_2O (650 mL) under magnetic stirring and maintained at 4 °C for one night to precipitate a maximum of chlorophylls. The resulting solution was successively extracted with $CHCl_3$, EtOAc, and *n*-butanol, respectively. The organic phases were dried with anhydrous Na_2SO_4 and filtered by common filter paper and concentrated in vacuum up to 35 °C to obtain the following extracts: $CHCl_3$ (2 g), EtOAc (7 g), and *n*-butanol (40 g). All the extracts were kept in the freezer until they were used.

Animals and treatments

Young adult female Wistar albino rats (*n*, 36; average age, 48 ± 5 days) weighing 150 ± 18 g were used in this study. They were obtained from the Laboratory Animal Research Unit of OMUDEHAM (Animal Ethics Committee, Ondokuz Mayis University, Samsun, Turkey). Ondokuz Mayis University was given ethical approval (OMUHAYDEK: B-30-2-ODM-0-20-09-00-050-04-32, 2015, 18th of April 2015). Animals were housed in cages, minted in an air-conditioned room at 22 to 26 °C with 12-h light and dark cycle and fed on standard rat pellets with free access to food and water ad libitum. Rats were acclimatized to the laboratory environment for 2 weeks, prior to the commencement of the study. The European Community Directive (86/609/EEC) and National Rules on animal care have been followed.

PCP (20 mg/kg body weight) and/or plant extract (50 mg/kg b.w.) were administered daily by gavages for 2 weeks. The vehicle used in the administration of the compounds is the distilled water (d.w) which given as 20 mg extract/0.5 mL

d.w/200 g b.w and 0.8 mg PCP/0.5 mL d.w/200 g b.w. Vitamin E was given intraperitoneally as a positive control at the dose of 100 mg/kg b.w. The doses used of extract and vitamin E were selected according to in vivo experiments, and studies carried out in our laboratory on the activity of different plant extracts on xenobiotic-induced toxicity in the liver (Amrani et al. 2017; Djebbari et al. 2017; Zama et al. 2007). Poison dose was selected based on acute PCP toxicity studies in wild animals and rats (Agha et al. 2013; Wang et al. 2001; Sai-kato et al. 1995; Villena et al. 1992). The LD50 (mg/kg) ranged from 80 to 120 in adult rats (St. Omer and Gadusek, 1987). The LD50 (mg/kg) ranged from 80 to 120 in adult rats (St. Omer and Gadusek 1987). Rats were divided at random into six groups:

- Group I (n = 6) was served as a control
- Group II (n = 6) was orally administered PCP 20 mg/kg b.w for 2 weeks (p.o.).
- Group III (n = 6) was orally administered plant extract 50 mg/kg b.w. (EXT50) for 2 weeks (p.o.)
- Group IV (n = 6) was orally treated by plant extract and PCP (EXT50 + PCP) for 2 weeks (p.o.)
- Group V (n = 6) administered daily by Vitamin E (VITE) 100 mg/kg b.w. by intraperitoneal injection for 2 weeks
- Group VI (n = 6) treated by Vitamin E 100 mg/kg b.w. and PCP 20 mg/kg b.w. (VITE + PCP) for 2 weeks

Preparation of homogenate tissue:

Animals were not fed overnight at the end of 2 weeks and were immediately perfused with 0.9% NaCl (+4 °C) to immediately remove blood due to the diurnal variation. Blood collection is performed from the portal vein into heparin tubes. The rats are anesthetized with an injection of ketamine hydrochloride (1 mL/100 mg) and xylazine (1 mL/23.32 mg). The rat's livers were removed and then homogenized in cold KCl 1, 15% to make a 20% homogenate. Centrifugation at 3000 rpm for 15 min at + 4 °C is important to separate the supernatant. These supernatants were used for analyses of all antioxidant enzymes and kept in a refrigerator at − 80 °C for further analysis. The protein content in the supernatant was determined calorimetrically as described in by Lowry et al. 1951 and measured at 660 nm. Bovine serum albumin (BSA) was used as a standard.

In vivo assays

Lipid peroxidation assay (MDA measurement):

Lipid peroxidation progression is resolute in the supernatant of all homogenates. It was evaluated by measuring the formation of thiobarbituric acid reactive substances (TBARS) via the colorimetric method of Uchiyama and Mihara (1978). In this experiment, 3 mL of 1% phosphoric acid and 1 mL of 0.67 % thiobarbituric acid (TBA). The aqueous solution was added to 0.5 mL of homogenate (20%) and moved in the centrifuge tube. The mixture was left in a boiling water bath for 45 min, and then it was cooled to room temperature. Four milliliters of n-butanol was added to the mixture and mixed forcefully. Absorbance was read at 532 nm after separation of the n-butanol phase by centrifugation. MDA was employed singly as standard. The TBARs content in liver homogenate was given nmol MDA/mg protein.

Glutathione content measurement:

Reduced glutathione (GSH) content in each liver homogenate was tested chemically via Ellman's reagent as reported by Ellman 1959. The basis of this analysis is the reactive cleavage of (DTNB) 5,5′-dithiobis(2-nitrobenzoic acid) by sulfhydryl group and resulting in yellow color with great absorbance at 412 nm against reagent blank with not any homogenate. The GSH content in each liver homogenate was given nmol GSH/mg protein.

Evaluation of glutathione peroxidase (GPx) activity:

The determination of GPx activity in rat's liver homogenate was performed as designated by Flohé and Günzler 1984. In the existence of GSH, GPx causes the hydrogen peroxide (H_2O_2) reduction in the medium. Briefly, 0.2 mL of supernatant disjointed from liver homogenate was added to 0.4 mL GSH (0.1 mM) and 0.2 mL of Tris-buffered saline (TBS) solution (Tris 50 mM, containing NaCl 150 mM, pH 7.4), and then the tubes were mixed and incubated 5 min at 25 °C. 0.2 mL of H_2O_2 (1.3 mM) was added to the mixture. After 10 min, 1 mL trichloroacetic acid (1% TCA) was added in order to end the reaction. Then, the tubes were kept at 0–5 °C in an ice bath for 30 min. After centrifugation for 10 min at 3000 rpm, 0.48 mL of supernatant was taken and added to each tube. 2.2 mL TBS solution and 0.32 mL of Ellman's Reagent, 5,5′-Dithiobis-(2-Nitrobenzoic Acid) (DTNB) (1 mM) were added 5 min before the measurement of the optical density at 412 nm. The activity was given nmol GSH/mg protein.

Dosage of biochemical parameters

The clear serum supernatant was used for the analysis of aspartate aminotransferase (AST), alanine aminotransferase (ALT), cholesterol, and triglyceride parameters analyzed with Audit Diagnostics Instrument, Ireland. The assays were conducted using the kits obtained from the Faculty of Veterinary, Ondokuz Mayis University, Turkey.

Histological studies

Directly, after the sacrifice of rats' liver-samples were excised, rinsed with normal saline, and processed distinctly for histological interpretations. The material was preserved in the fixative (10% formalin) for 48 h, dried by serial ethanol cycles (70% to absolute), and placed in paraffin. The fragments were cut in 5 μm in thinness which colored with Harris hematoxylin and eosin, and then observed using light microscopy (Leica DM 1000, Germany) at the laboratory of anatomical and pathological cytology, University Hospital Center, Constantine, Algeria.

Statistical analysis

All investigates were achieved in every test in triplicate. Data are obtainable as mean ± standard deviation of the mean. Analyses were executed by the Statistical Package for the Social Sciences (SPSS) Software V.25 using Anova two-way analysis and assessed by Tukey test. The difference $p < 0.01$ is considered as significant variation while $p < 0.001$ mentioned the highly significant variation. The averages followed by different letters are significantly different according to the Tukey test ($p \leq 0.05$).

Results

Effects on MDA level

A significant ($p < 0.01$) variation was detected in MDA level in rats treated with PCP plus plant extract (50 mg/kg). Extract administration normalized the value of MDA level compared to PCP group. Vitamin E provided a significant protection against PCP-induced lipid peroxidation (Fig. 1).

Effects on GPx activity

Oxidative worry encouraged by PCP initiated a significant ($p < 0.001$) alteration in liver antioxidant resistance system as GPx level, compared to the control group. The glutathione peroxidase activity was significantly decreased ($p < 0.01$) in the liver of the PCP group. Co-treatment with plant extract and vitamin E led to an acceleration in the activity of the enzyme. After consumption of PCP with extract and vitamin E, the GPx activity was nearer to results for these factors in the control group (Fig. 2).

Effects on glutathione

A significant ($p < 0.001$) change in the GSH content was caused by the PCP treatment compared to the control. In the co-treatment with plant extract and vitamin E, the concentration of reduced glutathione was considerably augmented in the liver compared to the control (Fig. 3).

Effects on biochemical parameters

A significant augmentation ($p < 0.01$) in the serum AST and ALT activities was detected in the PCP group; when compared to the control group. Pre-administration of plant extract or vitamin E lowered the AST and ALT activities significantly ($p < 0.01$) compared to the PCP group alone. A maintaining of normal levels of serum transaminases was shown in the plant extract and vitamin E groups (Fig. 4).

A significant increase ($p < 0.01$) in the serum of cholesterol and triglyceride levels was showed in the PCP group compared to the control group. While in the co-treatment with plant extract or vitamin E, the cholesterol and triglyceride levels were significantly decreased in serum (Fig. 5).

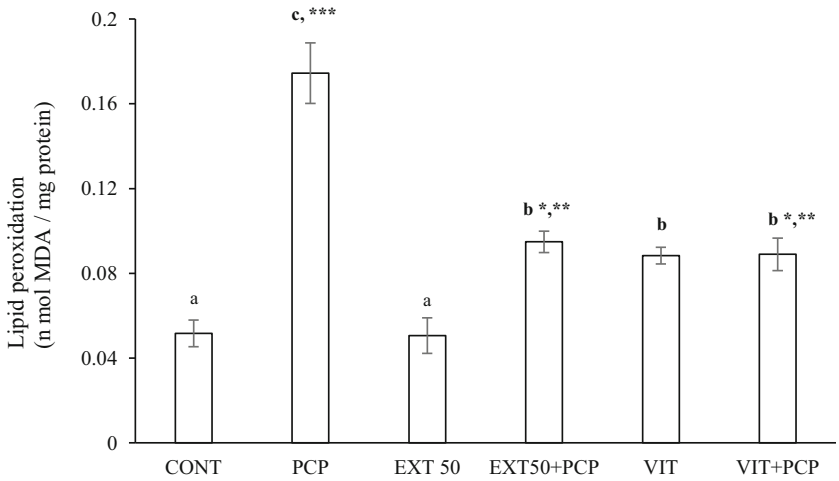


Fig. 1 Effect of PCP, *n*-butanol extract of *P. coronopifolia*, and vitamin E on lipid peroxidation (TBARs content) in liver homogenate. The averages followed by different letters (a–c) are significantly different according to the Tukey test ($p \leq 0.05$). *Compared to PCP group; ** $p < 0.01$ and *** $p < 0.001$, compared to control group. CONT, control group; EXT 50, plant extract at 50 mg/kg dose; VIT, vitamin E

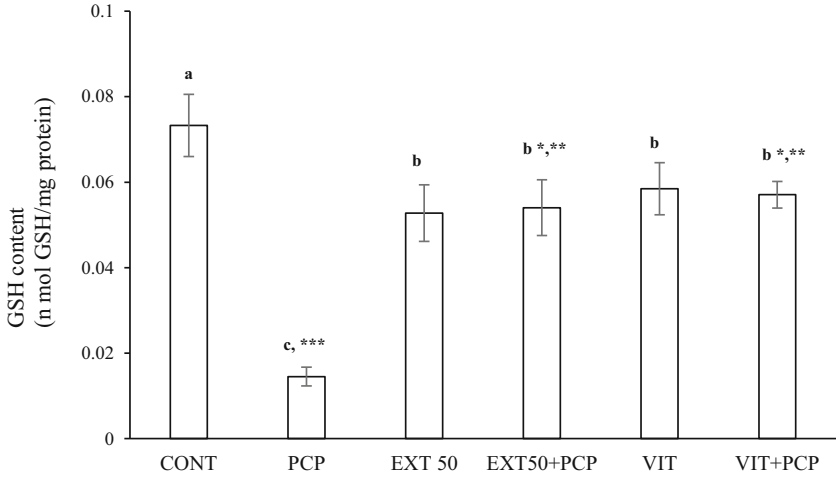


Fig 2 Effect of PCP, *n*-buthanol extract of *P. coronopifolia*, and vitamin E on GSH levels in liver homogenate. The averages followed by different letters (a–c) are significantly different according to the Tukey test ($p \leq 0.05$). *Compared to PCP group; ***$p < 0.001$, compared to control group. CONT, control group; EXT 50, plant extract at 50 mg/kg dose; VIT, vitamin E

Histological findings and results

Figure 6 a–d exhibit histological photomicrographs of liver-sections from control and treated groups. Histological photomicrographs of liver-sections from control exhibited a normal architecture with the central vein, polygonal hepatocytes with rounded nuclei (Fig. 6a). Histology of the liver treated with PCP illustrates a dilation and congestion of the centrilobular vein and sinusoids, pyknotic liver cells, binucleation, and ballooning of hepatocytes and necrosis (Fig. 6b). While the histology of the liver treated with plant extract or vitamin E before administration of PCP shows just edema of the centrilobular vein and less of dilation of sinusoids (Fig. 6c, d).

Discussion

Several in vivo methods complimenting antioxidant potency have been performed in this study. These methods have done to evaluate the protective effect of the *n*-butanol extract of *P. coronopifolia* on the maintenance of the antioxidant system against xenobiotics. This effect has been demonstrated via animal experimentation by inducing acute intoxication in adult rats by PCP.

Whereas, the ingesting of PCP for 2 weeks with a dose of 20 mg/kg in rats has caused damage to the electrical system in the hepatocyte membranes and a significant functional alteration in the livers. This hepatic dysfunction is demonstrated by the significant increase in MDA, one of the main products resulting from lipid peroxidation. Oxidative stress in the liver has been exacerbated by PCP since a significant decrease in GSH levels and an inhibition of GPx action compared to the control group. As well as, the examination of biochemical parameters in the serum of these rats recorded an increase in AST and ALT transaminases indicating lipid peroxidation at hepatocyte membranes. These changes in lipid profile have similarly proved by the increase of serum cholesterol and triglyceride levels in comparison with the control group. On the other hand, pretreatment of rats with the *n*-butanol extract of *P. coronopifolia* has shown that it is able to normalize the MDA level, to reduce the glutathione level and the GPx activity in liver homogenates. This extract is also able to decrease the AST and ALT transaminases activities and capable to decrease the cholesterol and triglyceride concentrations in the serum of rats poisoned by PCP.

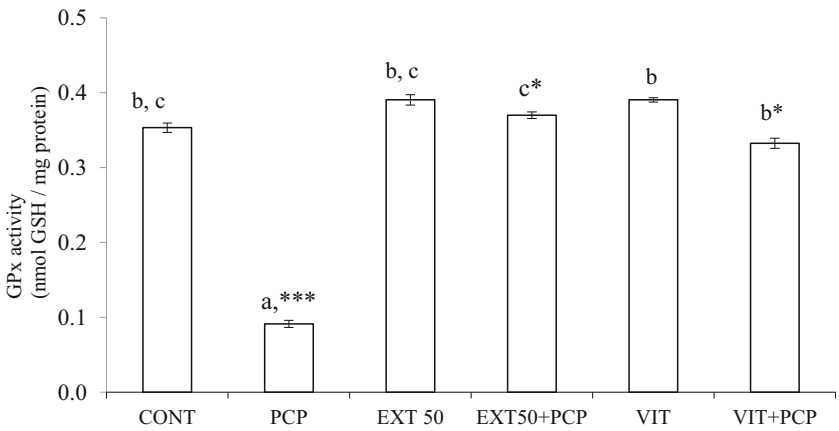


Fig 3 Effect of PCP, *n*-butanol extract of *P. coronopifolia*, and vitamin E on the antioxidant enzyme (GPx) in liver homogenate. The averages followed by different letters (a–c) are significantly different according to the Tukey test ($p \leq 0.05$). *Compared to PCP group; ***$p < 0.001$, compared to control group. CONT, control group; EXT 50, plant extract at 50 mg/kg dose; VIT vitamin E

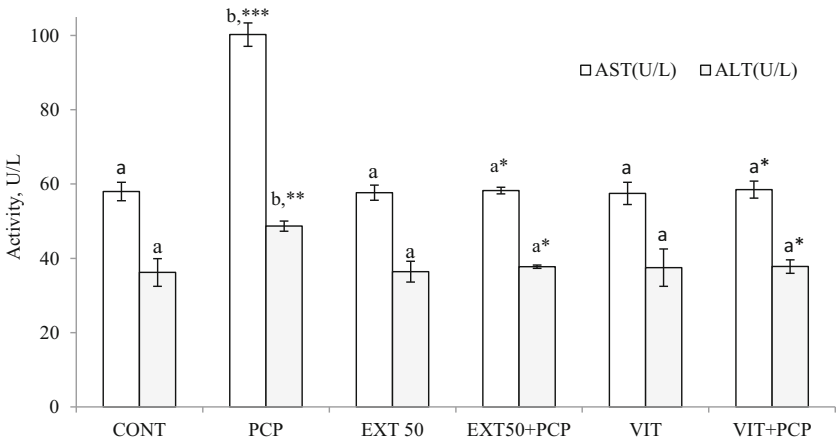


Fig 4 Effect of PCP, n-butanol extract of *P. coronopifolia*, and vitamin E in serum AST and ALT activities. The averages followed by different letters (a, b) are significantly different according to the Tukey test ($p \leq 0.05$). *Compared to PCP group; **$p < 0.01$ and ***$p < 0.001$, compared to control group. CONT, control group; EXT 50, plant extract at 50 mg/kg dose; VIT, vitamin E

Hepatic damage using pesticides persuaded lipid peroxidation commonly well-known and has been extra-evaluated in investigational assays to comprehend the cellular mechanisms overdue oxidative injury and estimate the beneficial potential of remedies and dietetic antioxidants (Basu 2003; Zama et al. 2007; Zhou et al. 2013). The obtainable human and animal findings specify that the metabolism of PCP does happen in the liver, via two ways: the conjugation to produce the glucuronide and the oxidative de-chlorinating to create tetrachlorohydroquinone (TCHQ) (Wang et al. 2001).

Previously researches recently reported that the metabolism of oral-PCP administration is able to progress over the quinols TCHQ and Cl_4CAT by microsomal cytochrome P_{450} enzymes (Fang et al. 2015). These TCHQ able to be oxidized through semiquinone intermediates (tetrachloro-1,2-semiquinone [Cl_4-1,2-SQ] and tetrachloro-1, 4-semiquinone [Cl_4-1,4-SQ]) into the correspondent quinones (tetrachloro-1,2-benzoquinone [Cl_4-1,2-BQ] and tetrachloro-1,4-benzoquinone [Cl_4-1,4-BQ]) (Carstens et al. 1990). Quinones and semiquinones are electrophilic and are able to be fixed with macromolecules in cells (Zhu and Shan 2009). The beginning of toxic properties was associated with the injury of the electrical system in membranes and a significant quantity of PCP binding to the membrane. That leads to the production of both superoxide anion and hydrogen peroxide at the very high rate which increases oxidative damage (Kan et al. 2015). This might well induce changes in enzymes involved in oxidative phosphorylation (Zhou et al. 2013).

In these experiments, the administration of 20 mg/kg of PCP for 15 days induced an increase of ROS as a result of stress disorder in the rats. Severe acute liver dysfunction and reflected modifications in the basic status of these organ revealed by an important escalation in the MDA levels as indicated in Fig. 1. It was caused by the interaction of oxygen radicals with polyunsaturated fatty acids. In accordance with our study, previous studies registered that pesticides (Umosen et al. 2012) and especially PCP increased MDA levels in various rat tissues compared to healthy models (Agha et al. 2013; Han et al. 2009). However, pretreatment with *n*-butanol extract of *P. coronopifolia* and vitamin E indicated its anti-lipoperoxidative effect owing to their antioxidant potent and free radical scavenging capacity over the bio-membrane restoration of liver parenchyma cells.

Furthermore, oxidative stress in the liver was aggravated by PCP since the increase in GSH levels, as shown in Fig. 2. This factor is considered as very important to make known

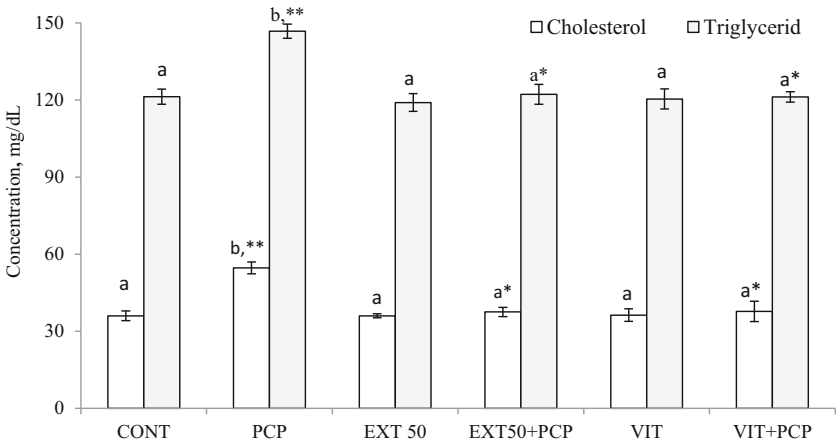
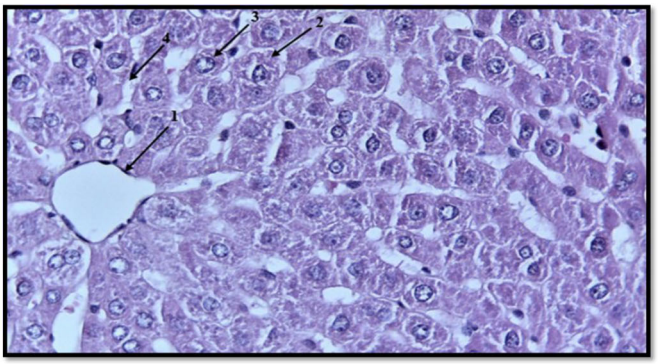
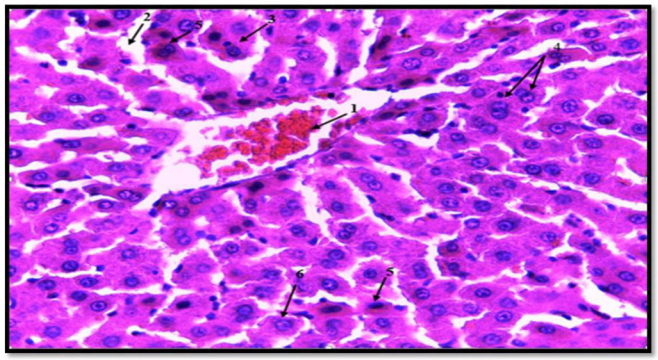


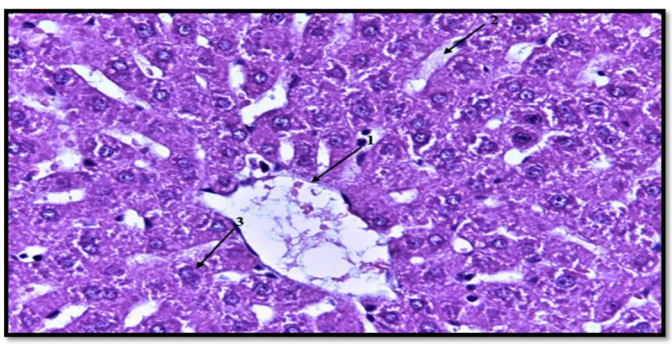
Fig 5 Effect of PCP, *n*-butanol extract of *P. coronopifolia*, and vitamin E in serum cholesterol and triglyceride levels. The averages followed by different letters (a, b) are significantly different according to the Tukey test ($p \leq 0.05$). *Compared to PCP group; **$p < 0.01$, compared to control group. CONT, control group; EXT 50, plant extract at 50 mg/kg dose; VIT, vitamin E



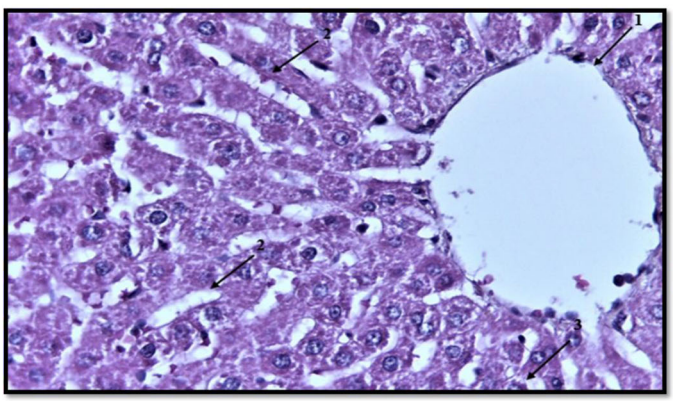
1: Normal architecture with central vein, 2: Polygonal hepatocytes, 3: A rounded nuclei, 4: Blood sinusoids. (X400).



1: Dilation and congestion of the centrilobular vein, 2: Dilation of sinusoids, 3: Pyknotic liver cells, 4: Binucleation of Hepatocytes, 5: Hepatocellular necrosis, 6: Ballooning of hepatocytes. (X400).



1: Dilation and edema of the centrilobular vein, 2: Dilation of sinusoids, 3: Binucleation of Hepatocytes. (X400).



1: Dilation of the centrilobular vein, 2: Dilation of sinusoids, 3: Ballooning and clarification of hepatocytes. (X400).

Fig 6 **a** Histology of normal control rat liver. (1) Normal architecture with central vein. (2) Polygonal hepatocytes. (3) A rounded nuclei. (4) Blood sinusoids (× 400). **b** Histology of liver treated with PCP. (1) Dilation and congestion of the centrilobular vein. (2) Dilation of sinusoids. (3) Pyknotic liver cells. (4) Binucleation of hepatocytes. (5) Hepatocellular necrosis. (6) Ballooning of hepatocytes (× 400). **c** Histology of liver treated with plant extract and PCP. (1) Dilation and edema of the centrilobular vein. (2) Dilation of sinusoids. (3) Binucleation of hepatocytes (× 400). **d** Histology of the liver treated with vitamin E and PCP. (1) Dilation of the centrilobular vein. (2) Dilation of sinusoids. (3) Ballooning and clarification of hepatocytes (× 400)

oxidative damage in the liver because the depleted GSH, in addition with GPx and GST (glutathione S-transferase), is responsible for the GSH redox cycles which keep the redox status of tissues against oxidative destruction persuaded by ROS and also guard their structural and regulatory proteins. The inhibition of GPx activity shown in Fig. 3 is related with the reduction of GSH, since the GSH substrate is used in the decomposition of H_2O_2 to water (Tiana et al. 1998) and the reduction of soluble hydrogen peroxide and alkyl peroxides by GPx (Bebe and Panemangalore 2003). In consequence, the reduction of GSH encouraged the formation of ROS and oxidative stress with a cascade of changes in the structural and possibly functional status of hepatic cells and organelle membranes. However, the pretreatment with plant extract or vitamin E qualified their biological importance in excluding ROS, which might disturb the typical role of cells.

Increased ALT enzymes shown in Fig. 4 were owed to the destruction of the hepatic structure. As soon as this enzyme is contained in the cytoplasm, it will be moved to the blood circulation after cellular change causing its augmented concentration. While, the increase of AST enzymes is being indicated that giving PCP to rats causes damage in both plasma and organelle membranes, which is supplemented by structural and functional adjustment of mitochondria. This result was agreed by other findings (Villena et al. 1992).

The *n*-butanol extract of *P. coronopifolia* regularized the ALT and AST activities. Normalization of ALT and AST levels suggests stabilization of the endoplasmic reticulum by pretreatment with this extract, leading to protein synthesis in the liver. Stimulation of protein synthesis has been advanced as a protective mechanism contributing to accelerate cell regeneration. This finding also suggests the hepatoprotective effect of the extract against the acute toxicity of PCP.

In addition, the present study mentions a significant increase in serum cholesterol and triglyceride levels in the PCP group compared to the control group as shown in Fig. 5. This finding also confirms that the administration of PCP caused changes in the lipid profile which lead to ballooning of hepatic cells. The progress of fatty liver is doing trough important mechanism which augmented substrate source for the esterification of fatty acids directly, motivated the esterification way, and also diminished the hepatic-triglyceride transfer like the very low-density lipoproteins. While, the precedents levels were decreased in serum by the pretreatment with plant extract or vitamin E due to their dominant inhibition of lipid peroxidation.

In the histopathological observations, histological sections of untreated rat's liver presented in Fig. 6a indicated a typical liver lobular architecture and cell arrangement. Although, Fig. 6b revealed entire liver structure damage of PCP-treated group with extreme dilation and congestion of the centrilobular vein, hepatocellular necrosis, dilation of sinusoids, pyknotic liver cells, bi-nucleation, and ballooning of hepatocytes. This finding concords with the results of Agha et al. (2013). Other studies confirmed that rats given oral dosages of 7–48 mg/kg/day of PCP presented hepatocellular necrosis, periportal fibrosis, and liver cells degenerating (Bernard et al. 2002).

Necrosis represents the dominant PCP-induced death pattern in different systems (Abhay and Sunanda 2015; Agha et al. 2013; Fernández et al. 2005; Chen et al. 2004; Wang et al. 2000; Villena et al. 1992). High-intensity oxidative stress can overwhelm the antioxidant potential and induce the opening of the transition port and mitochondrial permeability. Non-selective permeability of the inner membrane can lead to necrotic and apoptotic cell death (Lushchak 2011).

In the case of rats pretreated with plant extract and vitamin E, Fig. 6 c and d clearly confirm their protective effect. These findings concord with the results of Agha et al. (2013). The extract has shown a protective effect against lipid, electrical, and oxidative imbalance, as well as a hepatoprotective effect against acute toxicity induced by PCP in Wistar albino rats.

This ability to protect and preserve tissue may be provided by the large composition of polyphenolic content and flavonoids. The in vitro findings of Bekhouche et al. (2018) underlined the important composition of this extract of polyphenols which equal to 424.67 ± 4.03 μg gallic acid equivalent/mg of extract and also of flavonoids which equal to 347.67 ± 2.25 μg equivalents of quercetin/mg of extract. Many studies suggest that polyphenols have the capacity to regulate a variety of cellular and molecular processes by interaction with protein targets as enzymatic proteins, intracellular signaling proteins, nuclear receptors, etc (Amiot et al. 2009).

Polyphenols include phenolic acids and are rich in phenolic hydroxyl groups assigns antioxidant effects involving three processes: trapping the free radicals, inhibiting the generation of free radicals and anti-lipid peroxidation (Kolac et al. 2017). Phenolic acids are potential anti-liver damage compounds by protecting liver cells and preserving the integrity of the lysosomal membrane. This protection of the liver can be obtained by the following mechanisms: elimination of free radicals, inhibition of lipid peroxidation, inhibition of the expression of inflammatory cytokines, and improvement of the expression of lysosome-associated membrane protein 1 (LAMP1) which has significantly reduced by H_2O_2 attack (Wang et al. 2019; Yuan et al. 2015).

The study of Boussaha et al. (2015) has suggested that the antioxidant capacity of the aerial parts of *P. coronopifolia* is associated with the existence of flavonoid compounds such as taxifolin, rhamnazine, and the derivatives of the coffeeoylquinic acid determined in the ethyl acetate extract. Another recent study has suggested that the very potent anti-radical activity of this plant is due to the large amounts of certain major components such as γ-eudesmol, αeudesmol, and cis-nerolidol identified in essential oils of aerial parts of this plant (Hamdouch et al. 2017).

Moreover, our data assured that vitamin E has a considerable role in the evolution of acute hepatic toxicity and oxidative damage induced by PCP in the rat model as reported by Timbrell et al. (1995). Vitamin E has the capacity to inhibit lipid peroxidation, hepatocellular degeneration, necrosis, DNA injury, and lesions made in the extracellular matrix (Zhou et al. 1996). The *n*-butanol extract of *P. coronopifolia* can act as the vitamin E, as well as a radical chain terminator which converts reactive free radicals into stable, non-reactive products, and also as chemo-protectants against the oxidative damage induced in hepatocytes after ingesting PCP.

This in vivo study reconfirmed the in vitro findings of Bekhouche et al. (2018) which underlined that this plant extract has an important antioxidant potential compared to antioxidant standards and a high capacity of ROS scavenging, reducing power, metal chelation, DNA-damage protection, and anticancer activity against HeLa (human cervix carcinoma) cells at high concentration. These capacities were suggested to be offered by the high conception of polyphenols. Further studies are needed to better understand the possible mechanisms of action of this plant extract against PCP-induced toxicity in the liver.

Conclusion

This study can be shown as scientific support suggesting the hepatoprotective effect of the *n*-butanol extract of *P. coronopifolia* against PCP intoxication, which has been further confirmed by significant antioxidant activities and histopathological studies. The overdoses of PCP cause structural and functional alterations in the liver.

On the basis of the results obtained from this work, *P. coronopifolia* plant extract can be a prospective foundation of ordinary antioxidants and may provide a positive level of healthiness safety against oxidative damage induced by PCP.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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