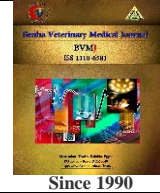




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### Original Paper

## Propolis alleviated Cisplatin-induced hepatorenal oxidative stress and apoptosis

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### ABSTRACT

The present research was planned to evaluate the protective effect of propolis water extract against cisplatin hepato-renal toxicity. Twenty-four male albino rats were divided into four equal groups. G1 (control) administered water only; G2 (propolis) administered (100 mg extract of propolis/kg body weight) orally by stomach tube at a once-daily dose for 14 days; G3 (cisplatin) administered once intraperitoneally (7.5 mg cisplatin/kg body weight) at day 10 of the experiment; and G4 (cisplatin+propolis) administered like G2 and G3. The serum biochemical analysis in the cisplatin group revealed an increase in AST, ALT, ALP, cholesterol, triglyceride, LDL-cholesterol, urea, and creatinine concentrations and a decrease in total protein and albumin concentrations. Moreover, cisplatin-treated rats significantly increased MDA levels and decreased SOD and CAT levels in hepatic and renal tissue compared to the control group. Administration of propolis (100 mg extract of propolis/kg.b.wt.) orally by stomach tube one hour before the second drug, which is once intraperitoneal (7.5 mg cisplatin/kg.b.wt.), at day 10 of the experiment (day number ten of the experiment, which continued for 14 days), reversed induced hepatorenal damage of the cisplatin group by exerting antioxidant and anti-inflammatory effects. Histopathological and immuno-histochemical examinations showed the hepatorenal damage induced by cisplatin was attenuated by propolis. In conclusion, propolis may help to lessen the hepatorenal damage induced by cisplatin via its antioxidant and antiapoptotic activities.

## 1. INTRODUCTION

Cisplatin is a potent anti-cancer drug that causes multi-organ toxicity, particularly in the liver and kidneys. Cisplatin, furthermore, causes inflammation and apoptosis (Eltamany et al., 2021). The precise method by which cisplatin induces cell death is still mostly unclear. Cisplatin is commonly assumed to generate intrastrand or interstrand crosslinks with purine bases on DNA strands. Crosslinking changes the ways that DNA is repaired, stops the making of a good DNA replication template, and speeds up cell-cycle arrest, which kills cells (Anbar et al., 2022). After infusion, cisplatin diffuses quickly into multiple organs with fast absorption, with a peak of blood plasma concentration after 5 minutes (Kireeva et al., 2020). Cisplatin is biotransformed by the cytochrome P450 (CYP450) enzyme complex (Abd Rashid et al., 2021).

Because the kidney is the primary excretory pathway for cisplatin, renal parenchyma accumulates it more than other tissues. An increase in cisplatin levels in the kidney leads to nephrotoxicity. It is thought that the negative nephrotoxic effects of cisplatin in renal tissue are caused by platinum buildup. Cisplatin accumulation stimulates the generation of TNF-alpha and ROS, hence increasing inflammation, oxidative stress, vascular damage, and apoptotic pathways

(Mcsweeney et al., 2021). Cisplatin causes oxidative damage to the liver; hepatotoxicity happens by inducing functional and structural mitochondrial injury, followed by apoptosis. Also, pro-inflammatory genes such as COX-2 and nitric oxide synthase play an important role in the mechanism inducing cisplatin hepatotoxicity (El-Gizawy et al., 2020).

Propolis is one of the natural resinous products produced by honey bees. Propolis water extract has been approved for the treatment of many diseases due to its antioxidant and anti-inflammatory properties. Rapidly upgrade the desire to use some natural products to treat many diseases. Propolis water extract is considered the best choice due to its very low cost in comparison to its value (Khaled et al., 2022). Propolis water extract from bee products is a promising major source of phenolic compounds with very strong antioxidant activity (Laaroussi et al., 2021).

This study was carried out to evaluate the protective effect of propolis against hepato-renal toxicity of Cisplatin by assessing serum biochemical hepato-renal biomarkers, antioxidant parameters, and histopathological and immunohistochemical changes in the liver and kidney.

## 2. MATERIAL AND METHODS

### 2.1. Chemicals

Cisplatin is commercially available as Cisplatin (MYLAN®) [1 mg/ml] from El Ezaby Pharmaceuticals, Egypt. Ready-extracted propolis powder was purchased from Imtenan Company (Cairo, Egypt), and after that, the powder was boiled in water to prepare propolis water extract (Pujirahayu et al., 2014). All analytical kits were obtained from Bio Diagnostic Company, Cairo, Egypt.

### 2.2. Gas chromatography-mass spectrometry (GC-MS) analysis of propolis water extract

The chemical composition of propolis water extract samples was determined using a GC-TSQ mass spectrometer (Thermo Scientific, Austin, TX, USA) and a TG-5MS direct capillary column (30 m x 0.25 mm x 0.25 m film thickness). The column oven temperature was initially kept at 60 °C and then increased by 5 °C/min to 250 °C with a 2-minute hold before increasing to 300 °C with 30 °C/min. The injector temperature was kept constant at 270 °C. Helium was employed as a carrier gas at a (1 ml/min) constant flow rate. The solvent delay was 4 minutes, and diluted samples of 1 µL were automatically injected using an auto-sampler AS3000 linked with a GC in split mode. In full scan mode, EI mass spectra were acquired at 70 eV ionization voltages spanning the m/z 50–650 range. The ion supply and transfer lines were set at 200 and 280 °C, respectively. The components were identified by comparing their 1. (mass spectra with WILEY 09) and 2. (mass spectral database NIST14). The spectral library currently contains more than 1600 fragmentation spectra that come from 435 authentic standards of endogenous metabolites and lipids (Abd El-Kareem et al., 2016)

### 2.3. Animals:

The rats were obtained from the Veterinary Serum and Vaccine Research Institute in Abbasia, Cairo, Egypt. Their starting weight was 200–240 g, and they were fed animal feed, a normal meal comprising 0.85% phosphorus, 1.12% calcium, 0.35% magnesium, 25.3% crude protein, and 2.5 IU g<sup>-1</sup> vitamin D<sub>3</sub>.

The Ethical Committee of Benha University's Faculty of Veterinary Medicine in Egypt approved all procedures of this experiment (Ethical No. BUFVTM 21-02-23).

### 2.4. Experimental design:

Twenty-four rats were randomly divided into four equal groups. Group 1: Rats served as the control group, and they received daily injections of water (1 ml) through their stomach tubes. Group 2 rats were given a propolis water extract of 100mg per kg of body weight. Orally by using a stomach tube daily (Laaroussi et al., 2021) and (Mohamed et al., 2022). Group 3: rats in this group were administered (7.5 mg Cisplatin/kg.b.wt.) once I/P injection on day 10 (only one time at day number ten from the experiment) (Alshahrani et al., 2022). Group 4: rats in this group were administered a 100-mg extract of propolis/kg b.wt. orally daily and 7.5 mg of cisplatin/kg b.wt. once intraperitoneally on the 10<sup>th</sup> day. The

experiment continued for 14 consecutive days. During this period, rats in all groups were observed daily.

### 2.5. Serum collection and tissue sampling

Rats were anesthetized with isoflurane one day following the final treatment at the end of the experiment. Blood samples were taken from the rats via direct piercing of the heart. The blood was put in plain tubes, allowed to coagulate, and then centrifuged at 3000 rpm for 15 minutes. The sera collected were used for biochemical analysis. The rats were sacrificed after their blood was collected, and their liver and kidneys were immediately removed. The tissues around the separated organs were removed, and the organs were cleaned with a 0.9 percent sodium chloride solution. The tissues were then dried using filter paper before being submerged in ice-cold phosphate-buffered saline with 0.1 mM EDTA to remove coagulated blood. The isolated liver and kidney tissues were then split into two sections. The first part was kept at -2 °C to test the activities of malondialdehyde (MDA), superoxide dismutase (SOD), and catalase (CAT). The second part was kept in 10% neutral-buffered formalin for histopathological and immunohistochemical studies.

### 2.6. Serum biochemical analysis:

Serum ALT, AST, and ALP were determined according to Babson et al., (1966); while the total protein and albumin levels were determined following the protocol of Doumas et al. (1971) and Koller and Kaplan, (1984) respectively using (Bio Diagnostics Company, Egypt) kits. Serum urea kits were used (Bio Diagnostics Company, Egypt) following the protocol of Coulombe and Favreau, (1963) and creatinine were analyzed using the protocol of Bartels et al., (1972). Cholesterol, triglycerides, and HDL cholesterol concentrations were analyzed according to the of Friedewald, (1972). All these biochemical parameters were measured using commercial-ready kits from Bio Diagnostics Company, Egypt. Serum LDL-cholesterol, total cholesterol, HDL cholesterol, and Triglycerides determined using ready to use kits from the BIOMED company Egypt.

### 2.7. Determination of antioxidant markers

One gram of liver and kidney tissue was homogenized in 5 ml of phosphate buffer, pH 7.4, using an electrical homogenizer. Tissue homogenates were centrifuged at 105,000 g for 60 minutes at 4 °C (Hu et al., 2021). The resultant supernatants were split into aliquots and kept at -2 °C to assess oxidative stress and antioxidant status. The amount of MDA was estimated according to Ohkawa et al. (1979). SOD activity was measured following Nishikimi et al. (1972), and catalase (CAT) was calculated according to Aebi, (1984).

### 2.8 Histopathological analysis

Kidney and liver tissue specimens were collected from rats and fixed in 10% neutral buffered formalin. Then dehydrated in ethyl alcohol, cleared in xylol, and embedded in paraffin wax. Five µm tissue

paraffin sections were prepared and stained with H&E stain (Bancroft and Stevens, 2016). Histopathological changes were evaluated with Leica microscopes (CH9435, Microsystems, Switzerland).

### 2.9. Immunohistochemistry examination (IHC)

The IHC sections were treated with antibodies. Markers were identified with peroxidase and stained with diaminobenzidine to differentiate antigen-antibody complexes. inspection and photography at various magnifications using a Leica microscope (Leica Microsystems, Switzerland).

### 2.10. Statistical analysis

Statistical analysis was conducted using the SPSS software program, version 20.0 (SPSS Inc., Chicago, IL). Statistical comparisons between the groups were performed using a one-way analysis of variance (ANOVA) followed by a Duncan's multiple comparison test. The results were expressed as *P*-values  $\leq 0.05$ , which were considered statistically significant.

## 3. RESULTS

### 3.1. Gas chromatography-mass spectrometry (GC-MS) analysis of propolis water extract

GC-MS was performed to identify volatile substances and detect molecules found inside propolis (water extract). It is an excellent technique for the identification of volatile substances. (GC) is the separation method for the component, and (MS) is the detector for molecule detection. (RT) considers the number of most important molecules. (MF) is the most important matching factor for these molecules. The GC-MS analysis of propolis (Table 1, Figure 1) revealed the detailed presence of 20 different phytochemical constituents. The most abundant constituents are octadecene, eicosene, and erucic acid. Volatile oils, which are essential oils, are a mixture of triglycerides, cholesterol, phospholipids, and saturated fatty acids such as stearic acid, palmitic acid, unsaturated oleic acid, and linoleic acid.

Table 1: Chemical composition of propolis as identified by Gas Chromatography-Mass Spectrometry (GC-MS)

NO	Compound Name	Area %	RT	Molecular Formula
1	9-Octadecenoic acid, methyl ester	20.29	28.91	C19H36O2
2	trans-13-Octadecenoic acid, methyl ester	20.29	28.91	C19H36O2
3	cis-13-Octadecenoic acid, methyl ester	20.29	28.91	C19H36O2
4	11-Octadecenoic acid, methyl ester	20.29	28.92	C19H36O2
5	Linoleic Acid methyl ester	14.35	28.73	C19H34O2
6	9,12-Octadecadienoic acid (Z,Z)-,methyl ester	14.35	28.73	C19H34O2
7	Methyl9-cis,11-trans-octadecadienoate	14.35	28.73	C19H34O2
8	(Z)-(Z)-Docos-13-en-1-yl icos-11-enoates	13.28	39.61	C42H80O2
9	cis-13-Eicosenoic acid	8.50	34.85	C20H38O2
10	cis-11-Eicosenoic acid	8.50	34.85	C20H38O2
11	Erucic acid	8.50	34.85	C22H42O2
12	cis-10-Nonadecenoic acid	8.50	34.85	C19H36O2
13	(E)-13-Docosenoic acid	6.88	36.68	C22H42O2
14	Ethanol, 2-(9-octadecenyloxy)-, (Z)-	4.54	36.63	C20H40O2
15	Stearic Acid Methyl Ester	2.36	29.47	C19H38O2
16	Methyl stearate	2.36	29.47	C19H38O2
17	Octadecanoic Acid, Methyl Ester	2.36	29.47	C19H38O2
18	Palmitic Acid methyl ester	1.49	25.71	C17H34O2
19	Hexadecanoic acid, methyl ester	1.49	25.71	C17H34O2

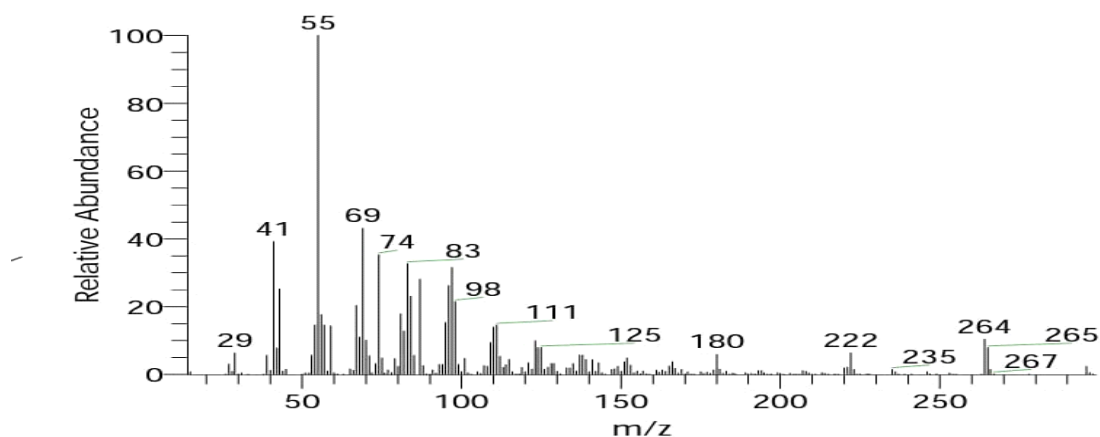


Fig 1: GC-MS analysis of propolis extract.

3.2. Effect of propolis water extract and/or cisplatin on serum biochemical parameters

Cisplatin (7.5 mg/kg b.wt.) injection on the 10th day of the experiment caused a significant increase in serum levels of ALT, AST, ALP, urea, creatinine, and a decrease in serum total protein (TP) and albumin activities compared with those in the control group. Supplementation of propolis (100 mg extract of propolis/kg b.wt.) together with cisplatin (7.5 mg cisplatin/kg b.wt.) for 14 consecutive days showed marvelously corrected results on liver (Figure 2) and renal biomarkers (Figure 2) ( $P \leq 0.05$ ).

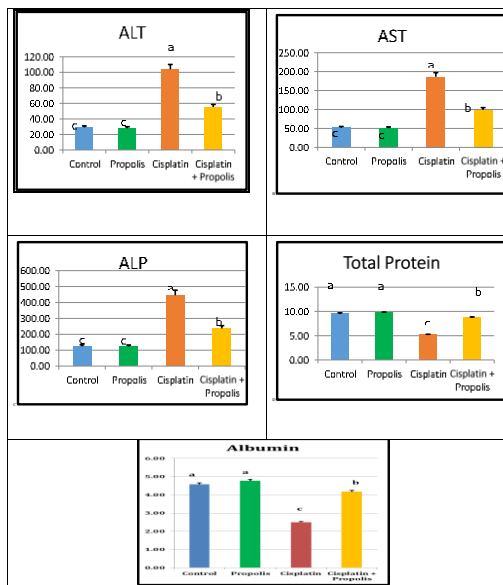


Fig 2: showing the effect of propolis water extract (100 mg extract of /kg.b.wt.) and/or Cisplatin (7.5 mg/kg.b.wt.) ALT (A) ,AST (B) ,ALP (C) , Total protein (D) and Albumin ( E) in serum of male albino rats

3.3. Effect of propolis water extract and/or cisplatin on lipid profile

Cisplatin (7.5 mg/kg b.wt.) injection on the 10th day of the study significantly elevated the lipid profile of cholesterol, triglyceride, and LDL levels and decreased HDL levels compared with those in the control group. Administration of propolis extract (100 mg/kg b.wt.) for 14 consecutive days showed a significant decline in cholesterol, triglyceride, LDL, and HDL levels and an increased HDL level compared with the cisplatin group, as shown in Fig. 3 ( $P \leq 0.05$ ).

3.4. Effect of propolis water extract and/or cisplatin on oxidative stress markers

Cisplatin (7.5 mg/kg b.wt.) injection on the 10th day of the experiment dramatically increased hepatic and renal MDA and decreased SOD and CAT in the liver and kidney tissues compared with the control group. On the other hand, propolis treatment in rats injected with cisplatin significantly attenuated hepatorenal oxidative stress and lipid peroxidation compared with the cisplatin group. ( $P \leq 0.05$ ; figure 4).

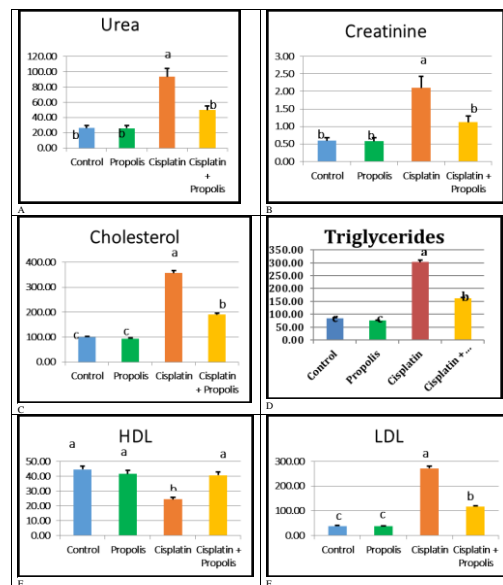


Fig 3: showing the effect of propolis water extract (100 mg extract of /kg.b.wt.) and/or Cisplatin (7.5 mg/kg.b.wt.) urea (A) , creatinine (B) , cholesterol (C) , Triglyceride (D) , HDL-c (E) and LDL-C (F) in serum of male albino rats.

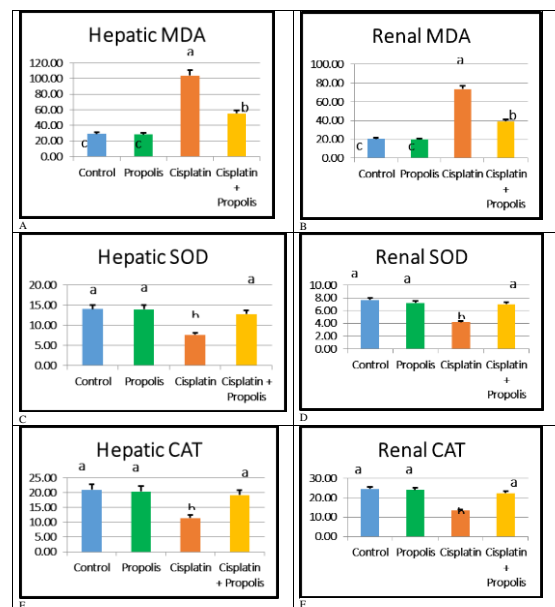


Fig 4: showing the effect of propolis water extract (100 mg extract of /kg.b.wt.) and/or Cisplatin (7.5 mg/kg.b.wt.) antioxidant parameters: hepatic MDA (A) ,renal MDA (B) , hepatic SOD (C) , renal SOD (D) , hepatic CAT (E) and renal CAT ( F) in male albino rats.

3.6. Histopathological findings:

Groups A (control) and B (propolis) showed normal histoarchitectures of both liver and kidneys. In contrast, group C (cisplatin) showed severe hepatic and renal injury. while group D (cisplatin + propolis) showed improvement along hepatic and renal tissues; most hepatocytes of the hepatic cords were seen as normal with enhanced along the renal cortex area where renal corpuscles were restored.

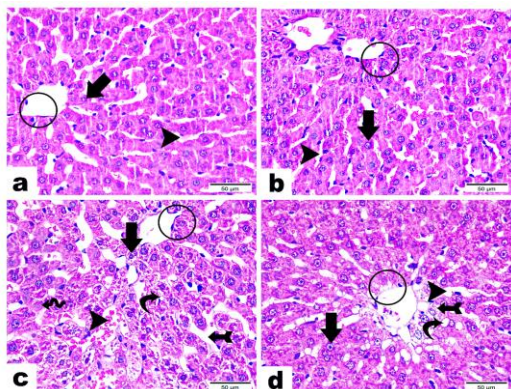


Figure (5): Photomicrographs displaying the effect of propolis on the hepatic tissue (central vein area) in the studied groups (H&E stain, x400 and scale bar= 50 $\mu$ m) as follow: Section from control group (a) and propolis group (b) revealing the standard structure of hepatic tissue with intact central vein endothelial lining (circle), well organized hepatic cords with normal light and vesicular nuclei of hepatocytes (arrows), hepatic sinusoids separated the hepatic cords and observed with its regular structure (arrowheads). Section from cisplatin group (c) highlighting severe hepatic injury with hydropic degeneration along hepatocytes (arrow), some hepatocytes appear with sever eosinophilic cytoplasm and karyolitic nuclei (wave arrow). Hepatic sinusoids exhibited dilatation with sever congestion (arrowhead). However, central vein presented with its normal structure (circle). Section from cisplatin and propolis group (d) showing noticeable improvement along hepatic tissue structure with few degenerations along central vein endothelial lining (circle), most hepatocytes of hepatic cords seen normal with light and vesicular form (arrow), while few ones appear with hydropic degeneration (arrow with tail), plus few amounts of microvascular steatosis were also seen (curvy arrow).

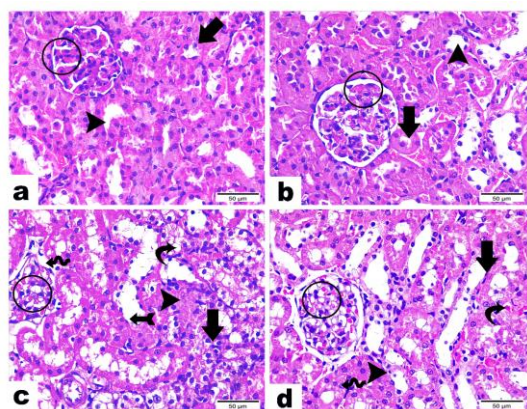


Figure (6): Photomicrographs exhibiting the effect of propolis on the kidney tissue (renal cortex area) among studied groups (H&E stain, x400 and scale bar= 50 $\mu$ m) as follow: Sections from control group (a) and propolis group (b) showing the normal histological structure of renal cortex area with normal glomerulus and renal corpuscle (cells) (circles), normal proximal convoluted tubules (arrows), as well as typical distal convoluted tubules (arrowheads). Section from cisplatin group (c) marking severe degenerative changes with atrophy of renal corpuscle, renal interstitial edema leading to dispersion between renal tubules (wave arrow). Some renal tubules loss their organization and showed pyknotic nuclei (arrow), with appearance of necrotic area (arrowhead), other renal tubules emerged dilated with squamous epithelial lining (arrow with tail), most tubules existed with high vacuolation (curvy arrow). Section from cisplatin and propolis group (d) displaying obvious enhancement along renal cortex area as renal corpuscle restore its regular organization but with some vacuolation and few RBCs (circle), some renal tubules appeared normal with light and vesicular lining cells (wave arrow), some still appeared with sever dilatation and squamous epithelial lining (arrow) and others spotted with deep basophilic apoptotic nuclei of lining cells (arrowhead), additionally congestion of the intertubular blood capillaries were also notice (curvy arrow).

### 3.8. Immunohistochemical Results:

Groups A and B there were scarce positive cytoplasmic reactivity with TNF- $\beta$  along hepatocytes, renal tubules, and renal corpuscles, but group C has high positive cytoplasmic reactivity with TNF- $\beta$  along hepatic and renal cells. On the other hand, group D was moderately positive cytoplasmic reactive with TNF-alpha along hepatocytes, renal corpuscles, and tubules.

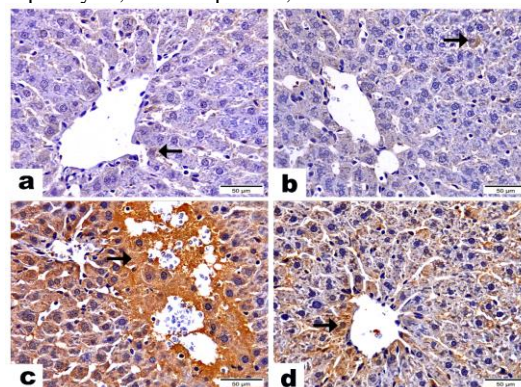


Figure (7): Photomicrographs displayed the expression of tumor necrosis factor alpha (TNF $\alpha$ ) along hepatic tissue (central vein area) between inspected groups (TNF $\alpha$ , x400, scale bar= 50 $\mu$ m) as follow: Sections from control group (a) and propolis group (b) showing very scarce positive cytoplasmic reactivity with TNF $\alpha$  along hepatocytes (arrows). Section from cisplatin group (c) showing high positive cytoplasmic reactivity with TNF $\alpha$  along hepatocytes (arrow). Section cisplatin and propolis group (d) displaying moderate positive cytoplasmic reactivity with TNF $\alpha$  along hepatocytes (arrow).

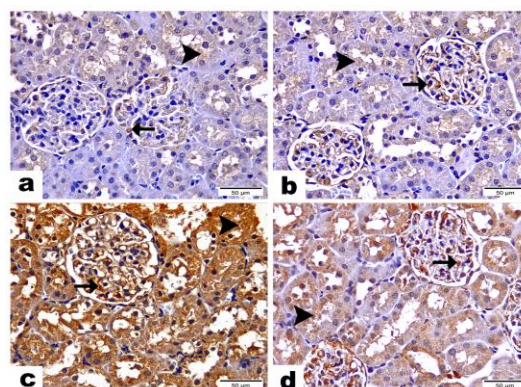


Figure (8): Photomicrographs demonstrating the expression of tumor necrosis factor alpha (TNF $\alpha$ ) along kidney tissue (renal cortex area) between inspected groups (x400, scale bar= 50 $\mu$ m) as follow: Sections from control group (a) and propolis group (b) showing scarce positive cytoplasmic reaction along renal corpuscle (arrows) and renal tubules (arrowheads). Section from cisplatin group (c) demonstrating marked positive cytoplasmic reaction along renal corpuscle (arrow) and also along renal tubules (arrowhead). Section cisplatin and propolis group (d) exhibiting moderate positive cytoplasmic reaction along renal corpuscle (arrow) and renal tubules (arrowhead).

## 4. DISCUSSION

Numerous medicines and chemotherapies used to treat various tumors have been linked to varying toxicological characteristics. As a result, the current study sought to assess the effect of propolis extract on protecting against cisplatin-induced hepato-renal damage. It has been found that IP injection of cisplatin provokes hepato-renal damage with a significant

increase in the serum levels of AST, ALT, ALP, urea, and creatinine, similarly to that observed in the results of the previous study (Rix et al., 2020). Remarkable elevations in liver index (ALT, AST) and renal biomarkers (urea and creatinine) may be associated with liver and kidney injury. The hepatorenal cytotoxicity of cisplatin is caused by mitochondrial disruption and promotes ROS production via an interrupted respiratory chain. ROS production allows oxidative damage to biological components such as DNA, proteins, and lipids (Bekhit et al., 2023). Cisplatin showed a reduction in total protein and albumin in comparison with the normal group due to its cytotoxic effect on liver and immune competent cells such as B lymphocytes and plasma cells, which means immunotoxicity (Zein et al., 2023). Also, administration of cisplatin increased the lipid profile (LDL, triglyceride, and cholesterol) but decreased HDL. Our result was in agreement with Abuzinadah et al. (2020). According to Dasari et al. (2022), cisplatin induces apoptosis by modulating the signal transduction pathways of mitogen-activated protein kinase (MAPK and p53) and subsequently increasing cisplatin sensitivity (chemo-sensitivity).

Interestingly, cisplatin induced a dramatic change in hepatic and renal oxidative parameters, with increased MDA and decreased SOD and CAT. This result came in agreement with Hashim et al. (2022). Cisplatin is taken up into the cells via an organic transporter like copper, causing damage to the nuclear DNA and the production of ROS, which eventually leads to cell death as cisplatin causes increased phosphorylation of all three proteins of the MAPK family, which is phosphorylation of JNK 1/2, ERK1/2, and p38 in hepatic tissues (Bekhit et al., 2023).

Histopathological abnormalities in liver and renal tissue confirmed the previous findings. There was hepatocyte degeneration with nuclear condensation, sinusoidal dilatation in the liver with congestion, and glomerular deterioration in the kidney, which was consistent with (Prasad and Prasad et al., 2021).

The results of immunohistochemistry revealed that the examined liver and kidneys of rats injected with cisplatin showed the highest positive cytoplasmic reactivity with TNF along hepatocytes, along renal corpuscles, and along renal tubules. Cisplatin may cause apoptotic or non-apoptotic death in cells by attaching to the purine bases of DNA and causing ruptures in the strand of DNA repair pathways (Elsayed et al., 2022). Also, cisplatin's hepato-renal cytotoxicity may be generated by mitochondrial dysfunction, the generation of superoxide anion and hydroxyl radical with lipid peroxidation, which causes membrane lipid peroxidation and polyunsaturated fatty acid breakage, resulting in cellular damage (Awad et al., 2023).

Propolis samples were evaluated for GC-MS to identify volatile substances and detect molecules found inside propolis (water extract). Propolis powder GC-MS analysis revealed 12 of the most relative molecules. The first of the three-four matching factors for every relative molecule. GC-MS analysis revealed propolis contains these bioactive components (fatty acids): (1<sup>st</sup>) (9-

Octadecenoic acid, methyl ester) and (2<sup>nd</sup>) (cis-13-Eicosenoic acid), which are considered antimicrobials according to Zahara et al. (2022). Antimicrobial peptides are small molecular peptides that play a crucial role in the innate immunity of the host against different microorganisms (Zhang et al., 2021). Also, 9-octadecenoic acid and methyl ester are the main bioactive compounds that are characterized by potent antioxidant and anti-inflammatory activities, according to the discussion in Ho et al. (2022). (3<sup>rd</sup>): Erucic acid has a role as a drug carrier and has neuroprotective effects (Galanty et al., 2023); also, erucic acid is considered an omega-9 fatty acid (Kumar et al., 2022). Omega-9 fatty acids are a kind of monounsaturated fatty acid that may be found in both plant and animal sources. They are classified as semi-essential fatty acids. Propolis, according to GC-MS, is considered a healthier alternative to saturated animal fats and has various health advantages, including anti-inflammatory and anti-cancer properties (Farak et al., 2022). (4<sup>th</sup>) (Cis 11-eicosenoic), (5<sup>th</sup>) (palmitic), as a chemical composition of propolis, has physico-chemical characteristics as well as DNA protective, cytotoxic, and antitumor activities (Andonova et al., 2023). (cis-11 eicosenoic acid) is considered the predominant very-long-chain fatty acid (Sarvas et al., 2021). Saturated fatty acids like palmitic acid (Tomata et al., 2021). The medicinal roles of these fatty acids were studied for their ability to inhibit in vitro the cell growth of three human cancer cell lines (Batsika et al., 2021). Furthermore, other investigations showed that propolis water extract has high antioxidant power due to the presence of fatty acids (Xu et al., 2022).

The results of this study revealed that administration of propolis extract (100 mg/kg BW) to rats injected with cisplatin showed that propolis extract prevented changes caused by cisplatin and improved hepatic and renal functions. This effect of propolis as a honeybee product is due to its antioxidant and anti-inflammatory activities, which were similar to those of Hossain et al. (2022). This hepatorenoprotective effect of propolis was evidenced by the reverse and correct elevation of levels of ALT, AST, ALP, urea, and creatinine in the serum. On the other hand, normalize the level of total protein, albumin. In addition, propolis showed significant improvement in the activities of cholesterol, triglycerides, LDL, and HDL. Moreover, remarkable normalized changes in liver and kidney oxidative stress markers (CAT, MDA, and SOD) were also recorded. This result was agreed upon (Laaroussi et al., 2021; Khaled et al., 2022).

The histopathological results of the examined liver and kidneys of rats injected with cisplatin and treated with propolis showed improvement along the hepatic and renal tissue structures. Most hepatocytes were seen as normal. The hepatic sinusoid exhibited few dilatations. As the renal cortex restored its regular organization, some renal tubules appeared normal. Also, an immunohistochemical study displayed moderate positive cytoplasmic reactivity with TNF along hepatocytes and renal cells. Therefore, the administration of propolis improved all the previously

mentioned biochemical and histological alterations induced by cisplatin that came into contact with (Okail et al., 2020). This protective role of propolis against cisplatin may return to the presence of chemical components, as shown by gas chromatography.

Finally, the obtained results provided a chance to advance the use of bee products like propolis as promising medicinal sources (El Adham et al., 2022).

## 5. CONCLUSION

The current study concluded that propolis had a possible hepatoprotective effect against cisplatin-induced hepato-renal oxidative injuries. This effect was evidenced by the significantly renormalized serum liver and kidney biochemical parameters and the re-corrected hepatorenal oxidative stress markers. The potential protective effect of propolis against oxidative stress could be attributed to its alteration of the apoptotic pathway and consequent upregulation of antioxidant enzymes. Thus, this study may contribute to the future hepatoprotective pharmacological application of propolis, thereby expanding its medicinal utility.

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