JJBS

Volume 12, Number 5,December 2019
ISSN 1995-6673
Pages 581 - 588

Jordan Journal of Biological Sciences

# An Evaluation of the Antioxidant Properties of Propolis against Fenvalerate-induced Hepatotoxicity in Wistar Rats

Wael M. Alamoudi*

*Department of Biology, Faculty of Applied Sciences, Umm Al-Qura University, Saudi Arabia.*

*Received February 2, 2019; Revised March 1, 2019; Accepted March 16, 2019*

**Abstract**

Fenvalerate is a synthetic pyrethroid pesticide used extensively in agriculture to protect a wide variety of crops. Propolis is a natural product produced by honey bees and is used widely in traditional medicines for its antioxidant properties. The present work evaluates the effects of fenvalerate administration on albino rats' liver and possible ameliorative roles played by propolis treatment. Fenvalerate treatment has induced histological changes in the liver of albino rats including the congestion of blood vessels, cytoplasmic vacuolization of the hepatocytes, necrosis and fatty degeneration. Biochemical analyses showed that fenvalerate caused a remarkable increase in the level of alanine aminotransferase (ALT), aspartate aminotransferase (AST), urea and creatinine, and a decrease in the total number of white blood cells (WBCs) and platelets. It also caused an increase in malondialdehyde (MDA) and depletion in the activity of the antioxidant enzymes, catalase (CAT), and superoxide dismutase (SOD) in the liver of rats. However, treating animals with fenvalerate followed by a propolis treatment led to an improvement in both histological and biochemical alterations induced by fenvalerate. The rats that were treated with fenv/propolis showed a dramatic improvement in the level of ALT, AST, urea and creatinine, WBCs, and platelets. Moreover, propolis was found to reduce the level of malondialdehyde and increase the activity level of antioxidant enzymes, SOD and CAT. In conclusion, propolis might be considered as a natural effective agent to eliminate the toxicity caused by fenvalerate, due to its antioxidant activity.

**Keywords:** Fenvalerate, Propolis, Hepatotoxicity, Transaminases, Antioxidants.

## 1. Introduction

The widespread utilization of insecticides in insect control has brought about the need for the evaluation of the hazards caused by such substances. Pyrethroids have been known as insecticides for many years, and are used as highly-active insecticides. The source of Pyrethroids is the flowers of the pyretherum plant *Chrysanthemum cinerariafolium* (Casida 1973). Due to the persistence of these insecticides in the environment, structures similar to Pyrethroids have been synthesized and proved to be effective against different insects (McEween and Stephenson, 1979). On the other hand, exposure to Pyrethroids was found to produce the serious side effects. It has been shown that animals exposed to these insecticides exhibited disturbances in their physiological activities in addition to other histopathological alterations (Amaravathi *et al.,* 2010; Giray *et al.,* 2011). Moreover, Fenvalerate is a cyanophenoxy-benzyl group of the synthetic pyrethroid pesticides used extensively in agriculture to protect a wide variety of crops; it also serves as an indoor pest control because of its high toxicity to insects (Kneko *et al.,* 1981).Moreover, it has been reported that fenvalerate can induce hepatotoxicity and alter the biochemical markers in experimental animals (Mani *et al*., 2004; Waheed *et al*., 2012) in addition to causing oxidative stress in rats (Prasanthi *et al.,* 2005).

Propolis is the substance that honey bees produce by mixing their own waxes with resins collected from plants. It has been widely used as a folk medicine since ancient times (Kumazawa 2018). Recently, propolis has gained popularity as healthy food in various parts of the world because it promotes health and prevents diseases (Gonzalez *et al*., 1994, Galeotti *et al*., 2018). Moreover, propolis has different biological effects and antibacterial activities including the inhibition of cell division, the inhibition of bacterial motility, disrupting the mechanism of cytoplasm cell membranes and cell walls, bacteriolysis, enzyme inactivation, and protein synthesis inhibition (AL-Ani *et al*., 2018; Sforcin *et al.,* 2000). It has also been reported as having anti-inflammatory (Khayyal *et al.,* 1993), anticarcinogenic (Bazo *et al*., 2002), antioxidative (Jasprica *et al*., 2007; Kanbur *et al*., 2009; Sobocanec *et al*., 2006) and hepatoprotective effects (Gonzalez *et al*., 1994). The components contained in propolis are more than 300, and these include sequiterpene quinines coumarins, phenolic aldehydes, steroids, polyphenol, inorganic compounds and amino acids (Khalil *et al.,* 2006). Earlier *in vivo* and *in vitro* studies indicated that there are numerous biological properties of propolis which include anti-inflammatory, immunomodulatory effects (Ivanovska *et al*., 1995), antitumor (Awale *et al*., 2008;

* Corresponding author e-mail: wmamoudi@uqu.edu.sa.

Ishihara *et al*., 2009; Aldemir *et al*., 2018), antioxidant (Jaiswal *et al.,* 1997) and free-radical scavenging effects (Pascual *et al.,* 1994). To the author's knowledge, literature and evidence on the protective effect of propolis on fenvalerate- induced alterations are not enough. Accordingly, the present investigation was designed to study the ameliorative effects and the protective role of propolis against fenvalerate hepatotoxicity in albino rats.

## 2. Materials and Methods

The experiment was performed according to the guidelines of the ethical committee of Umm Al-Qura University, Faculty of Applied Science, Makkah, KSA.

### *2.1. Chemicals*

Fenvalerate was purchased from a commercial agricultural market at Jeddah city KSA, propolis was purchased from the local health food market at Jeddah city, KSA. Alanine aminotransferase (ALT), aspartate aminotransferase (AST), urea and creatinine kits were obtained from (Sigma-Aldrich, St. Louis, MO, USA).

### *2.2. Propolis Extraction*

The Propolis extract was prepared in an ethanolic solution as described by Attalla and Ayman (2008). Propolis (10 g) was trimmed into small pieces and dissolved vigorously with 34.85 80 % (V/V) ethanol for forty-eight hours at 37 ± 1ºC. The resulted ethanolic solution was filtered through Whatman filter paper size No. 4. The obtained filtered solution was air-dried, and the extract was weighed and dissolved in a sterilized 0.9 % normal saline (PBS) for experimental processing.

### *2.3. Animals and Treatment*

Male Wistar rats, aged three months and weighing (120 ± 5 g), were obtained from the animal house of King Abdel Aziz University, Jeddah, Saudi Arabia. They were housed and maintained under standard laboratory conditions (24 ± 2 ºC, natural light-dark cycle) in well-aerated chambers, and were given free access to drinking water and commercial pellet diet. The animals were acclimated in laboratory conditions for one week before treatment. The animals were randomly divided into four groups with ten rats each.

Group I: (G1) Control group: animals were injected intraperitoneally (i.p.) with a buffer saline solution (0.9 % PBS).

Group II: (G2) Positive control: The animals were injected intraperitoneally (i.p.) with propolis at a dose of 100 mg/kg b.w. (Boutabet *et al*., 2011).

Group III: (G3) The animals were injected intraperitoneally (i.p.) with 5 mg/kg of fenvalerate (1/10 $LD_{50}$) dissolved in PBS, three times a week for four weeks. (Boutabet *et al.,* 2011).

Group IV: (G4) The animals were injected intraperitoneally (i.p.) with fenvalerate (5 mg/kg) followed by propolis (100 mg/kg) after twenty-four hours of the fenvalerate treatment, three times a week for four weeks (Boutabet *et al.,* 2011).

### *2.4. Tissue Preparation for Histopathological and Histochemical Studies*

At the end of the fourth week of treatment, all the rats were sacrificed by decapitation, and the liver pieces of each group were collected separately to be subjected to the histopathological, histochemical, and biochemical examinations. For the histopathological examination, small pieces of the liver were fixed in 10 % neutral formalin for twenty-four hours. Fixed materials were then processed and embedded in paraffin blocks. Sections that were 5 µm thick were cut and stained with Ehrlich's haematoxylin and eosin. For the histopathological examination, slides were later stained with Ehrlich's haematoxylin and eosin, whereas for the histochemical examination, the liver specimens were fixed in Carnoy's fluid for Periodic acid Schiff's reaction to demonstrate the content and distribution of polysaccharides as described by Kiernan (1999). The total protein contents were detected using the mercury bromophenol blue method as described by Pearse (1972).

### *2.5. Hematological and Biochemical Assays*

#### *2.5.1. Hematological Profile*

Fresh blood samples were used for the measurement of the red blood cells count (RBCs), white blood cells count (WBCs), hemoglobin content (Hb g/dI), and total platelets counts. Blood was obtained from the tail veins of controls and the treated groups, and was collected separately using an electronic blood counter. Differential WBCs were measured from blood smears on day 0 and weeks 1, 2, 3, and 4, respectively.

#### *2.5.2. Biochemical Assays*

For the biochemical study, blood was also collected, and the serum was obtained by centrifugating the blood samples (1500 rpm for ten minutes at 4 ºC) and stored at -20º for biochemical analysis. The activity of ALT and AST enzymes were measured using a diagnostic kit (Diagnostic system, Medford, NY, USA) as described by Reitman and Frankel (1957). The urea and creatinine were determined in the serum using a diagnostic kit (Diagnostic system, Medford, NY, USA) as described by Trinder (1969). Fresh tissue samples of the rats' liver were homogenized in cold distilled water until a uniform suspension was obtained. The homogenate was centrifuged, and the clear supernatant was separated. The Catalase (CAT) activity was also measured according to the method adopted by Aebi (1983). The superoxide dismutase (SOD) activity was determined according to the method described by Minami and Yoshikawa (1979). Malondialdehyde (MDA) was determined according to the method used by (Ohkawa *et al*., 1979)

### *2.6. Statistical Analysis*

The results were expressed as mean ± SD of different groups ($n$ =10). The statistical differences between the mean values were evaluated by ANOVA. Data were analyzed using the computer program SPSS/ version 15.0. Values were considered statistically significant when $p < 0.05$.

## 3. Results

### 3.1. Histopathological Observations

The histopathological examination of the livers of the control (G1) and the rats treated with propolis (G2) showed a normal hepatic architecture, and the cords of the hepatocytes radiated from the central vein to the periphery of the hepatic lobules. No histopathological alterations were observed from the normal and positive control groups (Figure 1A and B). However, the liver tissues of the rats treated with fenvalerate for four weeks (G3) showed apparent signs of pathological changes compared to the control group. The normal structural organization of the hepatic lobules was impaired with a clear hepatic degeneration; coagulative necrosis of many hepatocytes and blood vessels were congested (Figure 1C). Infiltration of inflammatory leucocytes was also noticed (Figure 1D). In addition, the hepatocytes revealed cytoplasmic vacuolation and the nuclei were pyknotic (Figure 2A). Moreover, impairment in the hepatocytes and necrosis signs with dense inflammatory cells and other debris were detected after four weeks of treatment with fenvalerate. In these tissues, a fatty degeneration composed of scattered fat droplets was clearly abundant (Figure 2B). On the other hand, in the animals treated with fenvalerate and propolis (G4), these histopathological changes became less. The hepatocytes regained normal architecture with normal achromatized central nuclei, and a noticeable reduction in the infiltration of inflammatory leucocytes was also noticed (arrow), (Figure 1E and F). Kupffer cells were activated and a large number of binucleated cells were noticed in these sections (arrows), (Figure 2C).

The histochemical observation of the livers of control rats (G1) showed a normal content of the total carbohydrates that appeared stained with a red or magenta colour with Schiff's reagent, and was not distributed in the cytoplasm of the hepatocytes, but appeared concentrated at one pole of the cells (Figure 3A). The examination of the livers of the rats treated with fenvalerate (G3) showed a diminution in their carbohydrates' content. The nuclei appeared negatively reacted (PAS-negative) confirming the absolute degradation of glycogen (Figure 3B). Figure 3C shows the remarkable increase in the carbohydrate content of the hepatocytes in the tissue of the rats treated with fenvalerate plus propolis (G4). Also, the total protein content of the hepatocytes of the control rats (G1) was positively reacted and stained with a blue color after being stained with a bromophenol blue. The protein content in the hepatocytes appeared to be distributed in the cytoplasm as fine granules (Figure 4A). Moreover, the chromatin bodies and nucleoli exhibiting deep coloration, Kupffer cells, and the endothelial lining cells of sinusoids showed a mild reactivity with a bromophenol blue. In addition, the walls of blood vessels revealed a strong stainability. The liver of the rats treated with fenvalerate (G3) showed a noticeable reduction in the total protein contents in the cytoplasm of the hepatocytes (Figure 4B). However, the liver cells of the rats treated with fenvalerate plus propolis regained a somewhat normal content of total protein (Figure 4C).

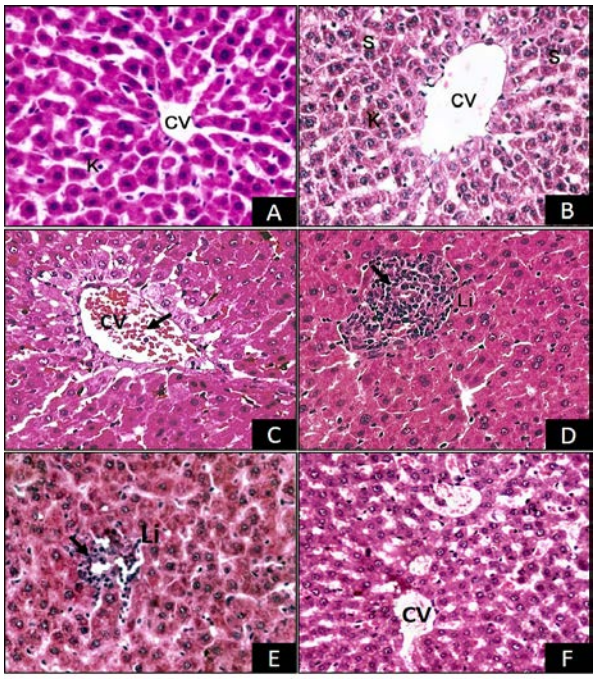


**Figure 1. (A)** Liver section of a control rat showing hepatic strands, kupffer cells (K), and central vein (CV). **(B)** Liver of a rat treated with propolis showing a normal hepatic architecture, kupffer cell (K) and sinusoids (S). **(C)** Liver of a rat treated with fenvalerate showing congestion of central vein (CV). **(D)** Liver of a rat treated with fenvalerate showing leucocytic infiltration (Li). **(E & F)** Sections of liver of rats treated with fenvalerate plus propolis showing a moderate degree of improvement in hepatocytes; few vacuolated hepatocytes exist with mild congestion in the central veins and lecocytic infiltration, (×400).

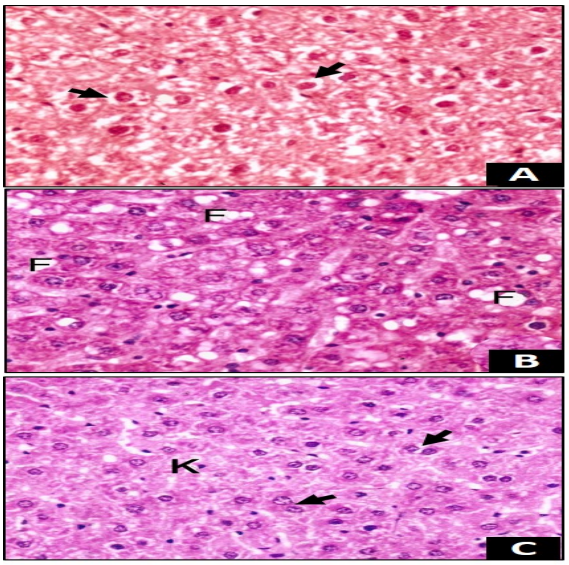


**Figure 2. (A)** Liver section of a rat treated with fenvalerate showing cytoplasmic vacuolizations of the hepatocytes and pyknotic nuclei (arrows). **(B)** Liver section of a treated rat showing fat droplets (F). **(C)** Liver section of a rat treated with fenvalerate and propolis showing an advanced degree of improvement in the hepatocytes which regained normal architecture with binucleated cells (arrow head) and activated kupffer cells (K), (×400).

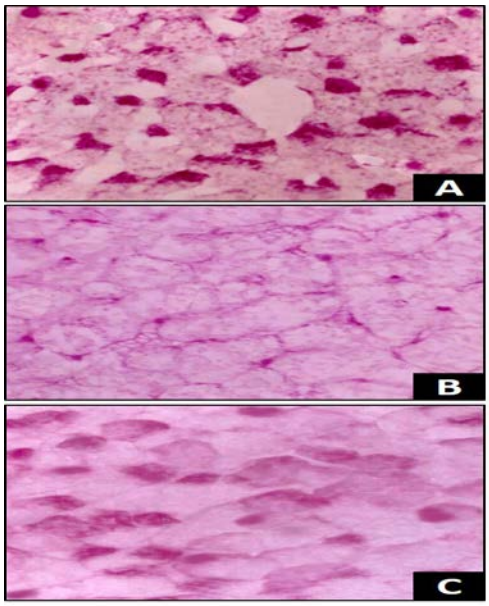


**Figure 3. (A)** Liver section of a control rat showing the distribution of carbohydrates in the cytoplasm of the hepatocytes. **(B)** Section of a rat treated with fenvalerate showing a noticeable decrease of carbohydrate in the hepatocytes. **(C)** Liver section of a rat treated with fenvalerate plus propolis showing a marked increase in the carbohydrate content in the hepatocytes, (×400).

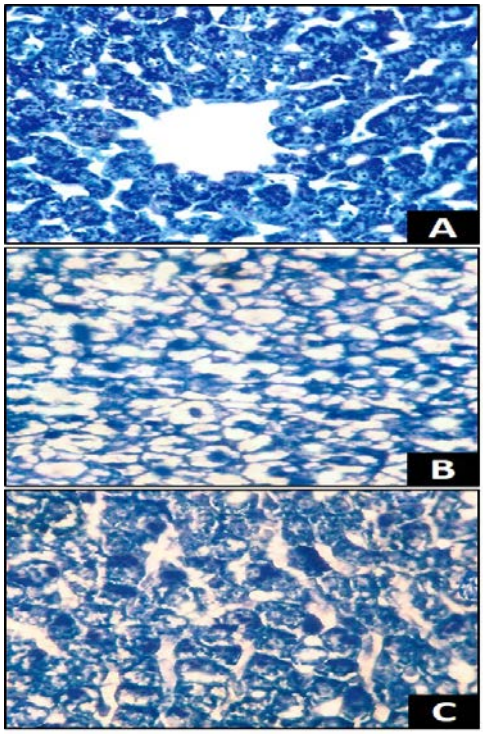


**Figure 4. (A)** Section of a liver showing the normal protein content of a control rat. **(B)** Section of liver showing a reduction of the protein content after treatment with fenvalerate. **(C)** Liver of a rat treated with fenvalerate plus propolis showing a noticeable increase in the protein content of hepatocytes, (×400).

### *3.2. Biochemical Parameters*

#### *3.2.1. Effect of Propolis on Fenvalerate-induced Changes in Serum Biochemical Parameters*

According to the results of the study, it was found that there is no significant change ($p \geq 0.05$) in the level of serum ALT, AST, urea and creatinine in the rats of the control (G1) and positive control (G2) groups. Treating rats with fenvalerate (G3) caused a significant increment ($p \leq 0.05$) in serum ALT, AST, urea and creatinine levels following a month of treatment in comparison with their levels in the serum of the (G1 and G2) groups. In other words, animals infused with fenvalerate alongside propolis (G4) demonstrated a significant decrease ($p \leq 0.05$) in the ALT, AST, urea and creatinine values compared with fenvalerate-infused group (G3). Importantly, they are still significantly higher ($p \leq 0.05$) compared with the control groups (G1 and G2) (Table. 1).

**Table 1.** Effect of propolis on fenvalerate-induced changes in serum biochemical parameters of liver in rats.

| | Control | Propolis | Fenvalerate | Fenv + Prop |
|---|---|---|---|---|
| **ALT (IU/L)** | 77 ± 0.4 | 72 ± 0.55 | 107 ± 1.7 | 81 ± 0.8 |
| **AST (IU/L)** | 115 ± 1.6 | 116.8 ± 4.1 | 194.6 ± 2.6 | 143.2 ± 0.9 |
| **Urea (mmol/L)** | 15.4 ± 0.6 | 17.3 ± 1.4 | 49.5 ± 0.8 | 28.9 ± 0.5 |
| **Creatinine(µmol/L)** | 1.4 ± 0.06 | 1.97 ± 0.31 | 3.16 ± 0.05 | 2.13 ± 0.06 |

Values are expressed as mean ± SD, $n$=10

#### *3.2.2. Effect of Propolis on Fenvalerate-induced Changes in Antioxidant Enzyme Levels*

Table 2 presents the biochemical results which showed that treatments on Malondialdehyde (MDA) (index of tissue lipid peroxidation), superoxide dismutase (SOD) and catalase (CAT) activity in the livers of animals examined after treatment for four weeks did not have any noticeable effects. There is no significant change ($p \geq 0.05$) in the enzyme levels of the control groups (G1 and G2). The MDA level was significantly increased ($p \leq 0.05$) in the rats infused with fenvalerate (G3), whereas the activity of SOD and CAT were significantly decreased ($p \leq 0.05$). However, treating the animals with fenvalerate alongside propolis (G4) demonstrated a significant decrease ($p \leq 0.05$) in the MDA level and significant increase in the SOD and CAT activity, compared to those of the control (G1) and positive control (G2) groups.

**Table 2.** Effect of propolis on fenvalerate-induced changes in MDA,CAT and SOD levels in the liver in rats.

| | Control | Propolis | Fenvalerate | Fenv + Prop |
|---|---|---|---|---|
| **MDA (u mole/g tissue)** | 133.5 ± 5.2 | 135 ± 6.3 | 192 ± 5.5 | 140.5 ± 2.3 |
| **SOD (Units/g tissue)** | 45.6 ± 2.8 | 44.7 ± 1.5 | 28.4 ± 2.5 | 36.5 ± 3.4 |
| **CAT (mole/min/g tissue)** | 0.33 ± 0.01 | 0.30 ± 0.02 | 0.12 ± 0.01 | 0.23 ± 0.01 |

Values are expressed as mean ± SD, $n$=10

### *3.3. Hematological Parameters*

#### *3.3.1. Total Number of White Blood Cells (WBCs)*

The present outcomes demonstrate that there is no significant change ($p \geq 0.05$) in the total number of WBCs in both control groups (G1 and G2) during the experiment period.

Nevertheless, there is a significant reduction ($p \leq 0.05$) in the aggregate number of WBCs in fenvalerate group (G3) and in the fenvalerate- and propolis-infused group (G4) in contrast with the (G1 and G2) groups until week

three and week four of treatment. The outcomes additionally demonstrate that there is no significant difference ($p \geq 0.05$) in the total number of WBCs in the fenvalerate and propolis group (G4) in contrast with the fenvalerate group (G3) until week three of treatment, (Table 3). During the fourth week of the experiment, the group treated with fenvalerate (G3) showed that they still have a significant drop in the total number of WBCs compared with the control groups (G1 and G2). Essentially, the group (G4) demonstrated a significant increase ($p \geq 0.05$) in the total number of WBCs in comparison with groups (G3 and G1). However, there was a nonsignificant increase ($p \geq 0.05$) in the total number of WBCs in comparison with the positive control (G2) (Table 3).

**Table. 3.**Effect of propolis on fenvalerate-induced changes in WBCs number in the serum of rats

| | Control | Propolis | Fenvalerate | Fenv + Prop |
|---|---|---|---|---|
| **Day 0** | 7.2 ± 0.7 | 7.5 ± 0.5 | 7.65 ± 0.35 | 7.73 ± 0.2 |
| **Week 1** | 7.12 ± 0.6 | 7.21± 0.7 | 3.51 ± 0.4 | 3.98 ± 0.9 |
| **Week 2** | 7.25 ± 0.5 | 7.0 ± 0.6 | 3.87 ± 0.8 | 4.17 ± 0.3 |
| **Week 3** | 7.20 ± 0.6 | 6.82 ± 0.35 | 3.69 ± 0.9 | 6.21 ± 1.14 |
| **Week 4** | 7.1 ± 1.12 | 7.7 ± 1.31 | 6.13 ± 0.4 | 8.9 ± 0.8 |

Values are expressed as mean ± SD, *n*=10

### *3.3.2. Total Number of Red Blood Cells and Hemoglobin Level*

The fndings demonstrate that there are no significant differnces ($p \geq 0.05$) in the aggregate number of RBCs and hemoglobin levels in the positive control (G2), control (G1) and the fenvalerate-infused group (G3). In contrast, the rats treated with fenvalerate plus propolis (G4) demonstrated a significant decrease ($p \leq 0.05$) in the aggregate of RBCs and Hb levels in comparison with the (G1, G2 and G3) groups respectively (Table 4).

**Table 4.** Effect of propolis on fenvalerate-induced changes in the RBCs number, Hb content and dential leucocytes in the serum of rats.

| | Control | Propolis | Fenvalerate | Fenv + Prop |
|---|---|---|---|---|
| **RBCs ($10^6$ /ml)** | 7.4 ± 0.5 | 7.6 ± 0.3 | 6.9 ± 0.4 | 5.8 ± 0.1 |
| **HGB (g/dl)** | 14.2 ± 1.0 | 13.1 ± 1.1 | 12.8 ± 0.6 | 10.6 ± 0.45 |
| **PLT ($10^3$ /ml)** | 346 ± 2.0 | 785 ± 2.5 | 575 ± 4.6 | 1235 ± 8.2 |
| **% Lymphocytes** | 68 | 66 | 63 | 7.1 |
| **% Monocytes** | 3.2 | 2.9 | 3.1 | 1.96 |
| **% Granulocytes** | 35.5 | 37.3 | 38.9 | 39.7 |

Values are expressed as mean ± SD, *n*=10

### *3.3.3. Total Number of Blood Platelets*

The findings demonstrate that the rats infused with propolis (G2) demonstrated a significant increase ($p \leq 0.05$) in the aggregate of platelets compared with the control group (G1). Furthermore, there was a significant increase ($p \leq 0.05$) in the platelets' number in the groups G1 and G2 compared with the rats treated with fenvalerate (G3). In addition, the rats treated with fenvalerate and propolis (G4) showed a significant increase ($p \leq 0.05$) in the platelets' counts compared with the control (G1) and the fenvalerate-infused (G3) groups (Table 4).

### *3.3.4. The Percentage of Differential Leucocytes*

Nonsignificant differences ($p \geq 0.05$) in the differential leucocytes in the control (G1, G2) and fenvalerate-infused (G3) groups were found in this study. Moreover, the group treated with fenvalerate plus propolis (G4) demonstrated a little increase in the percentage of lymphocytes and a decrease in the percentage of monocytes and granulocytes compared with the control (G1, G2) and the fenvalerate-infused (G3) groups (Table 4).

## 4. Discussion

Synthetic pyrethroids account for more than 30 % of insecticide use worldwide through household and agricultural applications. Nevertheless, exposure to these chemicals has been accompanied by several toxicities and health issues. This study is aimed at investigating the possible ameliorative effects of propolis after treatment with fenvalerate. The results indicate that the administration of fenvalerate to rats resulted in many histopathological, histochemical, and biochemical parameters including ALT, AST, urea and creatinine levels in the liver. The results show that there are no significant changes in the histological observation and biochemical analysis in the rats of the control group injected with PBS and the rats of the positive control group injected with propolis. The histopathological and histochemical observations of the rats treated with fenvalerate caused hepatotoxicity and noticeable changes in the hepatocytes architecture which appeared as hepatic degeneration and coagulative necrosis of many hepatocytes, cellular degeneration of some hepatocytes and cellular leucocytic infiltration.

The results also show a remarkable reduction in cytoplasmic carbohydrates and a degradation of protein contents in the hepatocytes of liver. This histological and histochemical change was significantly ameliorated following the propolis injection. These findings confirmed the toxic effects of fenvalerate on liver functions, and are similar with previous studies which indicate that the therapeutic dose of fenvalerate may cause liver toxicity in freshwater fish *Cirrhinus mrigala* and zebrafish (Velmurugan *et al*., 2007; Han *et al*., 2017). It has been reported that fenvalerate causes histopathological damage to the liver tissues, and alters the architecture of the organ cells in female rats (Jayachitra *et al*., 2016). In agreement with this result, Amaravathi has reported that treating rats with fenvalerate caused degenerative changes in the liver, haemorrhages, and mild fatty changes, infiltration of mono nuclear cells, and proliferation of bile duct (Kanbur *et al*., 2009). The inhalation of fenvalerate resulted in liver necrosis and fatty degeneration in rats (Mani *et al*., 2004). It was also observed that fenvalerate caused histopathological changes in the liver of rats, such as the degeneration and proliferation of hepatocytes forming acinar and pseudoglandular patterns (Ali, 2013). On the other hand, treatment with propolis could ameliorate the side effects caused by fenvalerate treatment, even though it could not return the changes to the normal levels. It has been reported by Ates *et al*., (2006) and Bhadauria *et al*., (2007) that the major compound, caffeic acid phenethyl ester (CAPE), found in the propolis may be responsible for the regulation of the antioxidant enzymes, the inhibition of lipid peroxidation, and the reduction of hepatic damage.

The propolis extract was shown to have a protective effect on hepatocytes against carbon tetrachloride (CCl)-induced injuries *in vitro* and *in vivo* (El-Khatib *et al.*, 2002 and Mahran *et al*., 1996). Kolankaya has reported that the treatment with propolis significantly prevented the release of transaminases, and significantly enhanced protein towards control, suggesting its hepatoprotective potential (Kolankaya *et al*., 2002).

In the current study, a significant increase in the levels of ALT, AST, urea, and creatinine in the serum of fenvalerate-injected rats was noticed compared with the normal and positive control groups. Also, a marked reduction in the level of SOD and CAT was observed in the fenvalerate-treated rats, while the MDA level has significantly increased compared with the control group. However, rats that were treated with propolis along with fenvalerate showed a significant decrease in the level of ALT, AST, urea, creatinine, and MDA, while the activities of SOD and CAT were significantly increased compared to the fenvalerate-treated rats; they were still higher than the control groups. The effects of fenvalerate on ALT and AST had been recorded by many researchers. It has been found that fenvalerate caused a significant increase in the activities of hepatic transaminases, ALP and LDH. (Prasanthi *et al*., 2005). It was confirmed in a previous study that the administration of fenvalerate to rats causes acceptance of toxicity to the liver as a result of the rise of liver-damage marker enzymes such as aspartate transaminase, alanine transaminase, Gamma Glutamyl transferase, and lactate dehydrogenase (Waheed and Mohamed, 2012). This study found that fenvalerate causes increment in the lipid peroxidation marker, MDA and the reduction of the antioxidant enzymes SOD and CAT. Similarly, it was maintained that the fenvalerate-treated rats demonstrated a reduced activity levels of SOD, CAT, GSHPx, and GSH in the liver homogenates, while the quantity of lipid peroxidation was high as a result of the increment in the level of MDA (Waheed and Mohamed, 2012). Thus, a rise in MDA and a decrease in the content of SOD and CAT might be identified with the oxidative pressure created in the hepatocytes of rats treated with fenvalerate.

Propolis is known to have antioxidant effects and free radical scavengers. It detoxifies a variety of free radicals and reactive oxygen intermediates (AL-Ani *et al*., 2018; Galeotti *et al.*, 2018). The antioxidant activities of propolis and its polyphenolic flavonoid components including flavonoids, aromatic acids, and their esters are related to their ability to chelate metal ions and scavenge singlet oxygen, superoxide anions, proxy radicals, hydroxyl radicals and peroxynitrite (Ferrali *et al*., 1997; AL-Ani *et al*., 2018, Galeotti *et al*., 2018). Also, propolis instigates the decrease of the expanded movement of AST, ALT, urea and creatinine values in the plasma of rats treated with $AlCl_3$ The present findings demonstrate that propolis diminished the lipid peroxidation conceivably by its antioxidant activity (Ferrali *et al.*, 2009). It was found that propolis also modulates the antioxidant enzymes and reduces lipid peroxidation processes in plasma, lungs, liver, and the brains of mice in a dose- and tissue-dependent manner (Shinohara *et al*., 2002). Luan stated in his work that propolis enhances the lipid profile, MDA and SOD activity in mice (Luan *et al*., 2000). Propolis can likewise lessen the levels of ROS, for example, H2O2 and NO, that may be in charge of its anti-inflammatory effects (Tan-No *et al*., 2006) in addition to the scavenging impact of propolis on free radicals delivered by liver in light of octylphenol toxicity (Saleh, 2012; Aldemir *et al*., 2018) revealed a discovery by Benguedouar who stated that propolis reduced superoxide anion radicals and restrained the lipid peroxidation in rats given doxorubicin and vinblastin (Benguedouar *et al*., 2008). The findings of the present study confirm the antioxidative properties of propolis and its ability to prevent damage induced by fenvalerate in albino rats.

The greater part of various classes of pesticides and chemotherapeutic medications have resistant suppressive symptoms, brought about by the division of the hematopoietic cells that are affected, leading to neutropenia and lymphopenia which weaken the immunity system (Saleh, 2012). The findings demonstrate that there is a nonsignificant difference in the aggregate number of RBCs or in the Hb level in the fenvalerate-infused rats. Moreover, rats that were infused with fenvalerate plus propolis demonstrated a significant reduction in their values in comparison with the control groups. Also, it was shown that the rats which were infused with propolis only or with propolis alongside fenvalerate demonstrated a significant increment in the aggregate number of the platelets in comparison with the normal and positive control groups. These findings demonstrate that propolis may display intense antiplatelet activities. Recent studies have demonstrated that propolis plays an essential part in improving oxidative stress, apoptosis and necrosis induced by chemotherapeutic drugs, pesticide chemicals, and heavy metal elements (Kocot *et al*., 2018). Moreover, propolis was found to have significant anti-inflammatory, antioxidant, antioroliferative, cytostatic, antibacterial properties due to the major compound, caffeic acid phenethyl ester (CAPE), that is extracted from propolis (Akyol *et al*., 2012; Kocot *et al*., 2018). Moreover, it has been demonstrated that propolis is a promising adjuvant with chemotherapy (Padmavathi, *et al*., 2006) and with immunization (Chu *et al.*, 2006)

## 5. Conclusion

In conclusion, the results presented in this investigation indicate that propolis may be considered as a potential natural product that can be used to ameliorate and prevent the adverse side effects, such as toxicity and immunosuppression, due to its anti-inflammatory and antioxidant properties.

## References

Aebi, H.E. 1983. Catalase. In: Bergmeyer HU., (Ed.), **Methods of Enzymatic Analysis,** Verlag Chemie, Weinhem, pp 273- 286.

Akyol S, Ginis Z, Armutcu F, Ozturk G, Yigitoglu MR and Akyol O. 2012. The potential usage of caffeic acid phenethyl ester (CAPE) against chemotherapy-induced and radiotherapy-induced toxicity. *Cell Biochem Funct* **30**: 438-443.

AL-Ani I, Zimmermann S, Reichling J and Wink M. 2018. Antimicrobial activities of European propolis collected from various geographic origins alone and in combination with antibiotics. *Medicines* **5(2):** 1-17.

Aldemir O, Yildirim HK and Sözmen EY. 2018. Antioxidant and anti-inflammatory effects of biotechnologically transformed propolis. *J Food Processing Preserv.,* **42(6):** 25-33.

Ali AM. 2013. The Effect of antioxidant nutrition against fenvalerate toxicity in rat liver (Histological and immunohistochemical studies). *Ann Rev Res Biol*., **3(4):** 636-648.

Amaravathi P, Srilatha CH, Kumari RV, Suresh Ramadevi V and Sujatha K. 2010. Histopathological changes in induced fenvalerate toxicity in rats and its amelioration with *Withania somnifera*. *Indian J Vet Pathol.,* **34(1):** 65-86.

Ates B, Dogru MI, Gul M, Erdogan A, Dogru AK, Yilmaz I and Esrefoglu M 2006. Protective role of caffeic acid phenethyl ester in the liver of rats exposed to cold stress. *Fund Clin Pharm* **20**: 283-289.

Attalla F and Ayman A. 2008. Protective effects of propolis against the amitraz hepatotoxicity in mice. *J Pharmacol Toxicol* **3**: 402-408.

Awale S, Li F, Onozuka H, Esumi H, Tezuka Y and Kadota S. 2008. Constituents of Brazilian red propolis and their preferential cytotoxic activity against human pancreatic PANC-1 cancer cell line in nutrient-deprived condition. *Bioorg Med Chem* **16**: 181-189.

Bazo AP, Rodrigues MA, Sforcin JM, de Camargo JL, Ribeiro LR and Salvadori DM. 2002. Protective action of propolis on the rat colon carcinogenesis. *Teratog Carcinog Mutagen* **22(3):** 183-194.

Benguedouar L, Boussenane HN, Kebsa W, Alyane M, Rouibah H and Lahoouel M. 2008. Efficiency of propolis extract against mitochondrial stress induced by antineoplastic agents (doxorubicin and vinblastin) in rats. *Indian J Exper Biol.,* **46**: 112-119.

Bhadauria M, Nirala SK and Shukla S. 2007. Propolis protects CYP2E1enzymatic activity and oxidative stress induced by carbon tetrachloride. *Mol Cell Biochem* 3**02**: 215-224.

Boutabet K, Kebsa W, Alyane M and Lahouel M. 2011. Polyphenolic fraction of Algerian propolis protects rat kidney against acute oxidative stress induced by doxorubicin. *Indian J Nephrol* **21**: 101-106.

Casida JE. 2006. **Pyrethrum, the Natural Insecticide**. Academic Press. London, N.Y, 329pp.

Chu WH. 2006. Adjuvant effect of propolis on immunization by inactivated Aeromonas hydrophila in carp (Carassius auratus gibelio). *Fish Shelfish Immunol* **21**: 113-117.

El-Khatib AS, Agha AM, Mahran LG and Khayyal MT. 2002. Prophylactic Effect of Aqueous Propolis Extract against Acute Experimental Hepatotoxicity *in vivo*. *Z Naturforsch C* **57(3-4):** 379-385.

Ferrali M, Signorini C, Caciotti B, Sugherini L, Ciccoli L, Giachetti D and Comporti M. 1997. Protection against oxidative damage of erythrocyte membrane by the flavonoid quercetin and its relation to iron chelating activity. *FEBS Lett* **416(2**): 123-129.

Galeotti F, Maccari F, Fachini A and Volpi N. 2018. Chemical composition and antioxidant activity of propolis prepared in different forms and in different solvents useful for finished products. *Food* **7(41):** 1-10.

Giray B and Hincal F. 2011. Fenvalerate induced hepatic oxidative stress in selenium- and/or iodine-deficient rats. *Hum Exp Toxicol* **30(10):** 1575-1583.

Gonzalez R, Rernirez. D and Rodriguez S. 1994. Hepatoprotective effects of propolis extract on paracetamol-induced liver damage in mice. *Phytother Res.,* **8(4)**: 229-232.

Han J, Ji C, Guo Y, Yan R, Hong T, Dou Y and An Y 2017. Mechanisms underlying melatonin-mediated prevention of fenvalerate-induced behavioral and oxidative toxicity in zebrafish. *J Toxicol Environ Health A* **80(23-24):** 1331-1341.

Ishihara M, Na K, Hashita M, Itoh Y and Suzui M. 2009. Growth inhibitory activity of ethanol extracts of Chinese and Brazilian propolis in four human colon carcinoma cell lines. *Oncol Rep* **22**: 349-354.

Ivanovska N, Nechev H, Stefanova Z, Bankova V and Popov S. 1995. Influence of cinammic acid on lymphatic proliferation, cytokine release and *Klebsiella* infection in mice. *Apidologie* **26**: 73-81.

Jaiswal AK, Venugopal R, Mucha J, Carothers AM and Grunberger D. 1997. Caffeic acid phenethyl ester stimulates human antioxidant response element-mediated expression of the NAD(P)H: Quinone oxidoreductase (NQO1) gene. *Cancer Res* **57:** 440-446.

Jasprica I, Mornar A, Debeljak Z, Smolcić-Bubalo A, Medić-Sarić M, Mayer L, Romić Z, Bućan K, Balog T, Sobocanec S and Sverko V. 2007. In vivo study of propolis supplementation effects on antioxidative status and red blood cells. *J Ethnopharmacol* **110(3):** 548-554.

Jayachitra G, Ponmanickam P, Vijaya Kumar J, Sevarkodiyone SP and Rajagopal T. 2016. The effects of fenvalerate on production of pheromone, hormone and histopathological changes in endocrine glands of female albino rats. *ANJAC J Sci* **15(2):** 61-74.

Kanbur M, Eraslan G and Silici S. 2009. Antioxidant effect of propolis against exposure to propetamphos in rats. *Ecotoxicol Environ Saf* **72(3):** 909-915.

Khalil ML. 2006. Biological activity of bee propolis in health and disease. *Asian Pac J Cancer Prev* **7(1):** 22-31.

Khayyal MT, el-Ghazaly MA and el-Khatib AS. 1993. Mechanisms involved in the antiinflammatory effect of propolis extract. *Drugs Exp Clin Res* **19(5):** 197-203.

Kneko H, Ohkawa H and Miyamaota J. 1981. Comparative metabolism of fenvalerate and the [2s, as]-isomer in rats and mice. *J. Pesticide Sci* **6**: 317-326.

Kiernan JA. 1999. **Histological and Histochemical Methods**. Theory and Practice, Pergamon Press, New York, 12(6), pp 479.

Kocot J, Kiełczykowska M, Luchowska-Kocot D, Kurzepa J and Musik I. 2018. Antioxidant potential of propolis, bee pollen, and royal jelly: possible medical application. *Oxidative Med Cellular Longevity* **3**: 1-29.

Kolankaya D, Selmanoglu G, Sorkun K and Salih B. 2002. Protective effects of Turkish propolis on alcohol-induced serum lipid changes and liver injury in male rats. *Food Chem.,* **78(2**): 213-217.

Kumazawa S. 2018. Bioactive compounds in bee propolis for drug discovery. *AIP Conference Proceedings* **1933(1):** 030001-5.

Luan J, Wang N and Tian L. 2000. Study on the pharmacologic effect of propolis. *J Chinese Medl Materials* **23(6):** 346-348.

Mahran LG, El-Khatib AS, Agha AM and Khayyal MT. 1996. The protective effect of aqueous propolis extract on isolated rat hepatocytes against carbon tetrachloride toxicity. *Drugs under Exper Clin Res.,* **22(6):** 309-316.

Mani U, Prasad AK, Sureshikumar V, Kumar P, Kewel LAL and Maji BK. 2004. Hepatotoxic alterations induced by subchronic exposure of rats to formulated fenvalerate (20% EC) by nose only inhalation. *Biomed Environ Sci* **17(3**) 309-314.

McEween FL and Stephenson GR. 1979. **The use and Significance of Pesticides in the Environment**. John Wiley and Sons. NY, pp538.

Minami M and Yoshikawa HA. 1979. Simplified assay method of superoxide dismutase activity for clinical use. *Clinica Chimica Acta* **92(3**): 337-342.

Newairy AA, Salama AF, Hussien HM and Yousef MI. 2009. Propolis alleviates aluminium-induced lipid peroxidation and biochemical parameters in male rats. *Food Chem Toxicol*., **47(6)**: 1093-1098.

Ohkawa H, Ohishi N and Yagi K. 1979. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. *Analytical Biochem.*, **95(2)**: 351-358.

Padmavathi R. Senthilnathan P, Chodon D and Sakthisekaran D. 2006. Therapeutic effect of paclitaxel and propolis on lipid peroxidation and antioxidant system in 7,12 dimethylbenz(a)anthracene-induced breast cancer in female Sprague Dawley rats. *Life Sci* **24**: 2820-2825.

Pearse AGE. 1972. **Histochemistry, Theoretical and Applied**, Churchill Livingstone, London. 23rd Edition.

Pascual C, Gonzalez R and Torricella RG. 1994. Scavenging action of propolis extract against oxygen radicals. *J Ethnopharmacol* **41**: 9-13.

Prasanthi K, Muralidhara and Rajini PS. 2005. Fenvalerate-induced oxidative damage in rat tissues and its attenuation by dietary sesame oil. *Food Chem Toxicol*., **43(2):** 299-306.

Reitman S and Frankel S. 1957. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am J Clin Pathol* **28(1):** 56-63.

Saleh EM. 2012. Antioxidant effect of aqueous extract of propolis on hepatotoxicity induced by octylphenol in male rats. *Acta Toxicol. Argent* **20(2):** 68-81.

Sforcin JM, Fernandes A, Lopes CAM, Bankova V and Funari SRC. 2000. Seasonal effect on Brazilian propolis antibacterial activity. *J Ethnopharmacol.*, **73(1-2**): 243–249.

Shinohara R, Ohta Y, Hayashi T and Ikeno T. 2002. Evaluation of antilipid peroxidative action of propolis ethanol extract. *Phytother Res.*, **16(4)**: 340-347.

Sobocanec S, Sverko V, Balog T, Saric A, Rusak G, Likic S, Kusic B, Katalinic V, Radic S and Marotti T. 2006. Oxidant/antioxidant properties of Croatian native propolis. *J Agric Food Chem.*, **54**: 8018-8026.

Tan-No K, Nakajima T, Shoji T, Nakagawasai O, Niijima F, Ishikawa M, Endo Y, Sato T Satoh S and Tadano T. 2006. Anti-inflammatory effect of propolis through inhibition of nitric oxide production on carrageenin-induced mouse paw edema. *Biol Pharma Bull.*, **29**: 96-99.

Trinder P. 1969. Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Ann Clin Biochem.*, **6**: 24-27.

Velmurugan B, Selvanayagam M, Cengiz EI and Unlu E. 2007. The effects of fenvalerate on different tissues of freshwater fish *Cirrhinus mrigala*. *J Environ Sci Health, Part B* **42(2):** 157-163.

Waheed MP and Mohamed HS. 2012. Fenvalerate induced hepatotoxicity and its amelioration by Quercetin. *Inter J Pharm Tech Res.*, **4(4):** 1391-1400.