

Ameliorative Effect of an Aqueous Mixture of Cinnamon and Ginger Tea Against Hepatotoxicity Induced in Rats by the Insecticide Diazinon: A Histopathological and Ultrastructural Study

Ihab K. Mohamed*

Department of Zoology, Faculty of Science, Ain Shams University, Cairo, Egypt

*Corresponding Author E-mail: ihabkmohamed@yahoo.com

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ABSTRACT

The present study aims to investigate the ameliorative role of an aqueous powders mixture of cinnamon and ginger tea against the insecticide diazinon-induced hepatotoxicity in adult male Sprague-Dawley white rats. The oral LD_{50} of diazinon “850.12 mg/kg body weight rat” was determined during the current investigation. The light microscope examination of the liver sections of the rats orally-treated with $1/10 LD_{50}$ doses of diazinon daily for the last 10 days of the 90 days of the experiment revealed damage of the liver architecture, cytoplasmic vacuolization of the hepatocytes, pyknosis or karyolysis of the liver cells’ nuclei, necrosis, blood congestion, widening of blood sinusoids, hypertrophy of Kupffer cells, infiltration of inflammatory cells. Also, examination of the liver tissue of the same group of rats by using the electron microscope showed presence of cytopathological alterations such as swelling of endoplasmic reticulum, degeneration of the mitochondria and Golgi apparatus. The blood sinusoids were markedly widened and contained residual bodies of the necrotic liver cells. The bile canaliculi exhibited deformation and their microvilli became very few and appeared as ghosts. All these previously-mentioned pathological changes were not noticed in the liver sections of the rats that were orally-given the cinnamon and ginger tea for 90 days, and orally-treated with $1/10 LD_{50}$ doses of diazinon daily for the last 10 days of the 90 days of the experiment. Therefore, the current results prove the potent ameliorative activity of the aqueous mixture of cinnamon and ginger tea against hepatotoxicity induced in rats by the insecticide diazinon.

Key words: organophosphates, induced hepatotoxicity, LD_{50} of Diazinon, cinnamon, ginger, hepatoprotective compounds, histopathology, ultrastructure, liver, rat.

INTRODUCTION

The citizens of the developing countries are surrounded by a lot of dangers which may non-intentionally affect their health. Many pollutants cause hepatotoxicity, when they are introduced to the environment with huge

quantities. Among these factors are the insecticides and pesticides that are being used to combat many agricultural pests. Unfortunately, such chemicals are very dangerous for humans.

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Generally, the citizens of the third world countries are more apt to dangers of the misuse and the introduction of such environmental pollutants in a large amount without well awareness of avoiding their dangerous harsh. In the developing world, exposure to the organophosphorous insecticides jeopardizes the lives of the citizens. The organophosphorous insecticide, diazinon is known to induce adverse effects in many organs of the different living species¹⁻⁶. Many authors reported that diazinon is a hepatotoxic compound for humans and animals⁷⁻¹⁰. Therefore, diazinon was selected in the present study in a trial to avoid its hepatotoxicity.

Nowadays, most of the active ingredients of the therapeutic drugs are extracted from natural products of plant origin¹¹. Recently, in the developing countries such as Egypt, there has been increasing awareness of the medicinal uses of natural products of plants such as cinnamon (*Cinnamomum zeylanicum*) that is considered as antioxidant¹²⁻¹⁴, anticancer^{15,16}, antimicrobial¹⁷⁻¹⁹ compounds. Other evidence suggested that ginger (*Zingiber officinale*) may be effective in the treatment of many diseases such as diabetes, gingivitis, toothache and asthma²⁰. Constituents from ginger also have anti-inflammatory, anti-oxidant and anti-carcinogenic properties^{21,22}.

It is believed that when multiple antioxidants are used in combination, they prevent vulnerability to other agents and synergistically pool their antioxidant properties, as the consumption of a mixture of garlic, ginger, and pepper help modulate oxidative stress caused by hypercholesterolemia in rats²³. Similar results were obtained by other authors who investigated the antioxidant effect of cinnamon and garlic extracts on some body organs of rats²⁴. Earlier studies dealt with the ameliorative role of some compounds such as vitamins E and C²⁵, black seed (*Nigella sativa*) extract²⁶, pomegranate molasses²⁷, tea (*Camellia sinensis*) and olive (*Olea europaea*)

leaves extract²⁸, selenium²⁹, β -carotene³⁰, and grapeseed oil³¹ against diazinon toxicity.

Bearing in mind these introductory remarks, the present work aims to investigate the impact of daily drinking of an aqueous powders mixture of cinnamon and ginger tea on the hepatotoxicity induced by the insecticide "diazinon".

MATERIALS AND METHODS

Administration and preparation of cinnamon and ginger tea:

According to earlier studies^{32,33} of the toxicity of cinnamon (*Cinnamomum zeylanicum*) and ginger (*Zingiber officinale*) extracts, there were no toxic effects of these natural compounds in rats that were orally-treated with low or high doses of their powders daily. In the present study, different low doses of an aqueous powders mixture of cinnamon and ginger tea were tested on male adult rats. Light and electron microscopical examinations revealed absence of signs of histopathological alteration in the liver sections of these treated rats. According to the popular herbal medicine, cinnamon and ginger are taken in doses around 1 g and 0.1 g daily respectively. In the present study, the doses given to the rats were calculated depending on such doses that are usually taken by humans and by using the equation given by Paget and Barnes³⁴; *i.e.* [70 x the human dose mg/kg x 0.018]. Therefore, the doses of the current experiment were 1.8 g of cinnamon, and 0.2 g of ginger powders. These amounts were added to 150 ml of distilled water then, boiled together. The tea was left to cool at the room temperature. Then, an amount of 1.5 ml/day (18 mg cinnamon and 2 mg ginger powders) of the tea was given orally to each rat. The tea was prepared freshly every day as long as the experiment proceeded. All components were purchased as clean powders in well-sealed small buckets from common shops for herbs and spices (on-shelf goods) in the Egyptian local market.

The insecticide diazinon:

Diazinon is an organophosphorous insecticide (O,O-diethyl-O-12-isopropyl-6-methyl-4-pyrimidinyl-thiophosphate); it was obtained – under the commercial name; diazinon 60% EC – from Kafr El-Zayat Pesticides & Chemicals Co., Kafr El-Zayat, Egypt. The diazinon is an emulsion with a concentration 60% of diazinon, which is the active component. In the present experiment, all doses of diazinon – to induce hepatotoxicity or determine its LD₅₀ in the adult male white rats – were diluted in corn oil and given orally to the rats using a stomach tube.

Determination of the oral LD₅₀ and administration of the diazinon:

In the current study, the oral LD₅₀ of the diazinon was determined according to the method of Reed and Muench³⁵. Accordingly, doses of diazinon were prepared in a geometric progression starting by a sublethal dose (LD₀) that kills 0% – 20% and ending by a lethal dose (LD₁₀₀) which kills 80% – 100% of the treated rats during the period of 24 hours. In these preliminary testes, it was found that the maximum sublethal dose was estimated at 317.6 mg/kg body weight. An increased factor of 1.2 was used for each successive dose till the LD₁₀₀ was obtained when 100% of rats were killed. This LD₁₀₀ was calculated at 1638.9 mg/kg body weight. One hundred male rats were divided randomly into 10 groups; each of 10 rats that were treated with the doses of diazinon during the mortality test (Table 1).

Therefore, in the present investigation, the rats were orally-treated with 1/10 of the LD₅₀ of diazinon in corn oil daily for the last 10 days of the 90 days of the experiment.

Used animals:

Adult male Sprague-Dawley white rats, *Rattus norvegicus*, were used. They were purchased from Helwan Farm for Experimental Animals, Cairo, Egypt. At the start of the experiment, their age ranged between 11 to 12 months and the weight of each rat was 180 ± 10 g. The animals were kept in cleaned and good aerated environment at room temperature (25° C ± 2°

C). The rats were fed standard food rodent pellets (Agricultural-Industrial Integration Company, Giza, Egypt) and drinking water *ad libitum*. This investigation was approved and conducted in accordance with the ethical guidelines set forth by the Department of Zoology, Faculty of Science, Ain-Shams University, Cairo, Egypt.

Experimental design:

After an acclimation period of one week, 24 rats were divided randomly into four groups; each of 6 rats. The experiment was carried out for 90 consecutive days. All doses of the experiment were orally-given to the rats by gavage.

Group I (negative-control group): Each animal received 1.5 ml/day of distilled water for 90 days; besides, each was given 0.5 ml/day of corn oil for the last 10 days of the 90 days of the experiment.

Group II (positive-control or tea group): Each rat was given 1.5 ml/day of the previously prepared tea that contains 18 mg cinnamon and 2 mg ginger powders for 90 days.

Group III (diazinon-treated group): Each rat received 1.5 ml/day of distilled water for 90 days; besides, each was treated with 1/10 LD₅₀ of diazinon in corn oil daily for the last 10 days of the 90 days of the experiment.

Group IV (tea and diazinon group): Each animal was given 1.5 ml/day of the previously prepared tea that contains 18 mg cinnamon and 2 mg ginger powders for 90 days; besides, each was treated with 1/10 LD₅₀ of diazinon in corn oil daily for the last 10 days of the 90 days of the experiment.

All animals were sacrificed under light ether anesthesia 24 hours after the last dose. Then, they were rapidly dissected.

Tissue processing for light microscopy:

For light microscopic examination, pieces of the liver tissue were fixed immediately after dissection in 10% formalin solution, then dehydrated in ascending series of ethyl alcohol, cleared in terpineol, and were then embedded in paraplast. Five µm-thick sections

were cut using American Optical microtome (AO-821), stained with haematoxylin and eosin, examined under light microscope, and finally digital photos were recorded.

Tissue processing for transmission electron microscopy:

The liver sections were rapidly fixed in 2.5% glutaraldehyde in 0.1M cacodylate buffer at 4 °C, for 4 hours. Cacodylate buffer (0.1M) was used for washing. Then, the sections were post-fixed in 1% osmium tetroxide for 2 hours, dehydrated in ascending series of ethyl alcohol, cleared in propylene oxide, and then embedded in epoxy resin. 70 nm-thick ultrathin sections were cut with an ultramicrotome (American Optical Co, USA), stained with uranyl acetate and lead citrate. Finally, the sections were examined and

photographed by using the transmission electron microscope (JEOL, 1200 EXII, Tokyo, Japan).

RESULTS

Determination of the oral LD₅₀ of the diazinon:

The oral LD₅₀ of the diazinon to the adult male Sprague-Dawley white rats, *Rattus norvegicus*, was determined according to the method of Reed and Muench [35]. Table “1” shows the doses that were used for the mortality test. To avoid the interference of accidental survival with deaths resulting from either resistance or susceptibility, the percentage of mortality at each dose, was calculated from the total number of survivals encountered at higher concentrations and these recorded with lower ones.

Table “1”: Calculation of oral LD₅₀ of diazinon in adult male Sprague-Dawley white rats *Rattus norvegicus*, using the Reed and Muench method³⁵

Doses mg/kg body weight rat	Number of rats	Died rats	Survived rats	Cumulative total			Percent mortality
				Died	Survived	Total	
317.6	10	-----	10	-----	58	58	0/58 = 0%
381.2	10	1	9	1	48	49	1/49 = 2%
457.4	10	1	9	2	39	41	2/41 = 5%
548.9	10	2	8	4	30	34	4/34 = 12%
658.6	10	3	7	7	22	29	7/29 = 24%
790.4	10	3	7	10	15	25	10/25 = 40% ^{**}
948.4^{***}	10	5	5	15	8	23	15/23 = 65% [*]
1138.1	10	8	2	23	3	26	23/26 = 88%
1365.8	10	9	1	32	1	33	32/33 = 97%
1638.9	10	10	-----	42	-----	42	42/42 = 100%

As difference of logarithms or proportionate distance = [(mortality at dilution next above 50%) – (50%)] / [(mortality next above 50%) – (mortality next below 50%)].

Therefore: Proportionate distance = [65^{*} – 50] / [65^{*} – 40^{**}] = 15 / 25 = 0.6

As log₁₀ 50% dilution (log₁₀ LD₅₀) = log₁₀ of dilution showing a mortality next above 50% – (proportionate distance X logarithm of dilution factor).

Therefore: log₁₀ LD₅₀ = log₁₀ (948.4^{***}) – (0.6 X log₁₀ 1.2) =
log₁₀ LD₅₀ = 2.97699 – (0.6 X 0.0792) = 2.97699 – 0.04751 = **2.92948**

Thus: Oral LD₅₀ of the diazinon for rats = antilog₁₀ (2.92948) = 850.12 mg/kg body weight

Light microscopic results**Group I (control group) and group II (tea group) – (Figs. 1a-b and 2a-b):**

The liver of rats of both groups I (-ve control group) and II (tea group) showed normal histological architecture. The liver is composed of thousands of hepatic lobules. Each lobule has prismatic polygonal shape and consists of liver cells. The liver cells form the hepatic strands that are radiating around the central veins of the hepatic lobules. All liver cells have normal polygonal appearance. Their basophilic-stained nuclei are large, spherical and almost centrally-located inside the granular cytoplasm. The liver cells are exposed on their sides to the blood flowing in the blood sinusoids of the hepatic lobules. The phagocytic cells of Von Kupffer are seen at the boundaries of most of the blood sinusoids. Among the lobules there are portal spaces that embody branches of the portal vein, the hepatic artery and the bile ductules. The lining endothelia of the central veins, the branches of the hepatic portal vein and the hepatic artery appear intact.

Group III (diazinon group) – (Figs. 3a-d):

The liver sections of the rats treated with 1/10 of the LD₅₀ of diazinon in corn oil, for the last 10 days of the 90-days of the experiment; revealed histopathological alterations in the liver tissue. These changes included loss of the liver architecture. The majority of the hepatocytes showed vacuolization and shrinkage of the cytoplasm. Pleomorphic nuclei of the liver cells were common. Some cells possessed pyknotic nuclei. Karyolysis was also observed in some hepatocytes. Some regions of the liver tissue exhibited complete necrosis that was clearly observed in many hepatic lobules. Many intervening hepatic blood sinusoids showed marked widening. Some areas of the liver lobules showed obliterated blood sinusoids, while a few sinusoids became very narrow and congested with blood. Most of Kupffer cells appeared hypertrophied. Some congested central veins were occasionally seen. The endothelium of

many central veins and the liver cells around them displayed noticeable degeneration. Examination of the liver sections of this group showed deformation of the portal triad, damage of the surrounding parenchymatous cells and loss of the hepatic architecture, as well. Many branches of the hepatic portal vein were markedly swollen and massively congested with blood. The cells of the limiting plate in the portal area – around the portal triad – were completely deformed and in some areas became very thick. Marked swelling of the branches of the hepatic arteries was also observed. No pathological signs were seen in the bile ductules. Many inflammatory cells infiltrated among the foci of the necrotic hepatocytes. Collectively, all these features of the liver of rats treated with diazinon showed moderate to severe signs of hepatotoxicity, which were diffusely distributed throughout the hepatic tissue.

Group IV (tea and diazinon group) – (Figs. 4a-c):

The protective effect of cinnamon and ginger tea against the induced hepatic histopathological alterations was proved by the histological examination of the liver sections of rats of group IV. The liver sections of such rats showed normal histological structure of the hepatic tissue. Normal parenchymal architecture with cords of polygonal hepatic cells was observed. The cytoplasm and the nuclei of the hepatocytes revealed no degenerative changes. Many blood sinusoids exhibited normal features, but only very few blood sinusoids showed a little wideness. Most of the Kupffer cells appeared normal in size. Some Kupffer cells occasionally exhibited a mild degree of hypertrophy. The central veins, the branches of the hepatic portal vein and the hepatic arteries were not congested, and the whole portal triads exhibited a normal appearance. The previously-mentioned characteristics of the liver tissue of the rats of this group were similar to those of the healthy liver of rats of the two control groups (I and II).

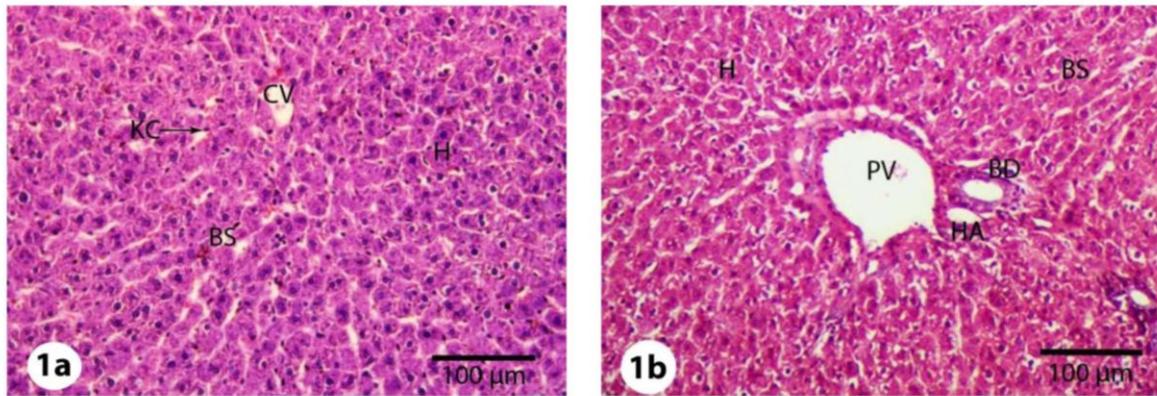


Fig. 1: Liver sections of untreated control rats of group I showing the strands of normal hepatocytes (H), the blood sinusoids (BS) and Kupffer cells (KC). The central vein (CV) is seen in **Fig. 1a**. A branch of the portal vein (PV), a branch of the hepatic artery (HA), and a bile ductule (BD) are noticed in **Fig. 1b**. Hx and E.

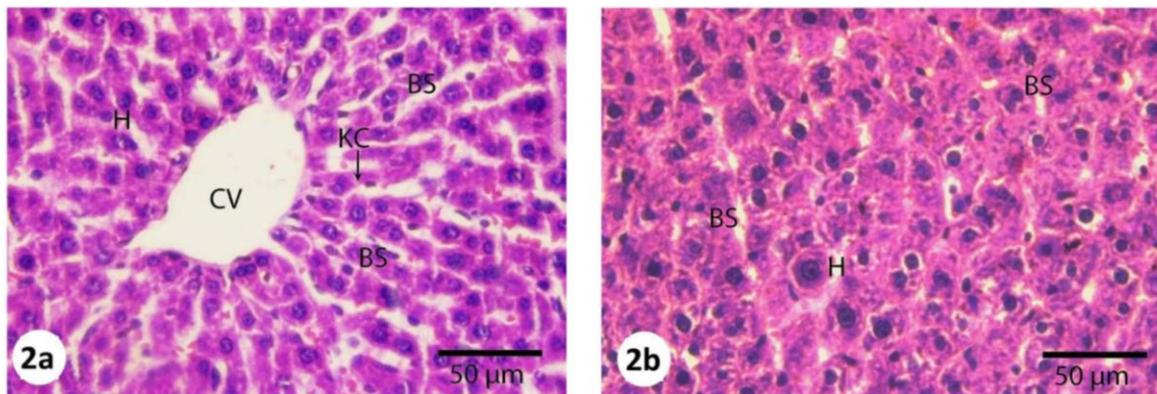


Fig. 2: Liver sections of rats of group II that were orally-given the herbal tea (18 mg of cinnamon and 2 mg of ginger powders) daily for 90 days. Notice the strands of normal hepatocytes (H) that alternate with the blood sinusoids (BS). The nuclei of the liver cells appear large, spherical and almost centrally-located. Normal Kupffer cells (KC) are also noticed. The central vein (CV) is seen in **Fig. 2a**. Hx and E.

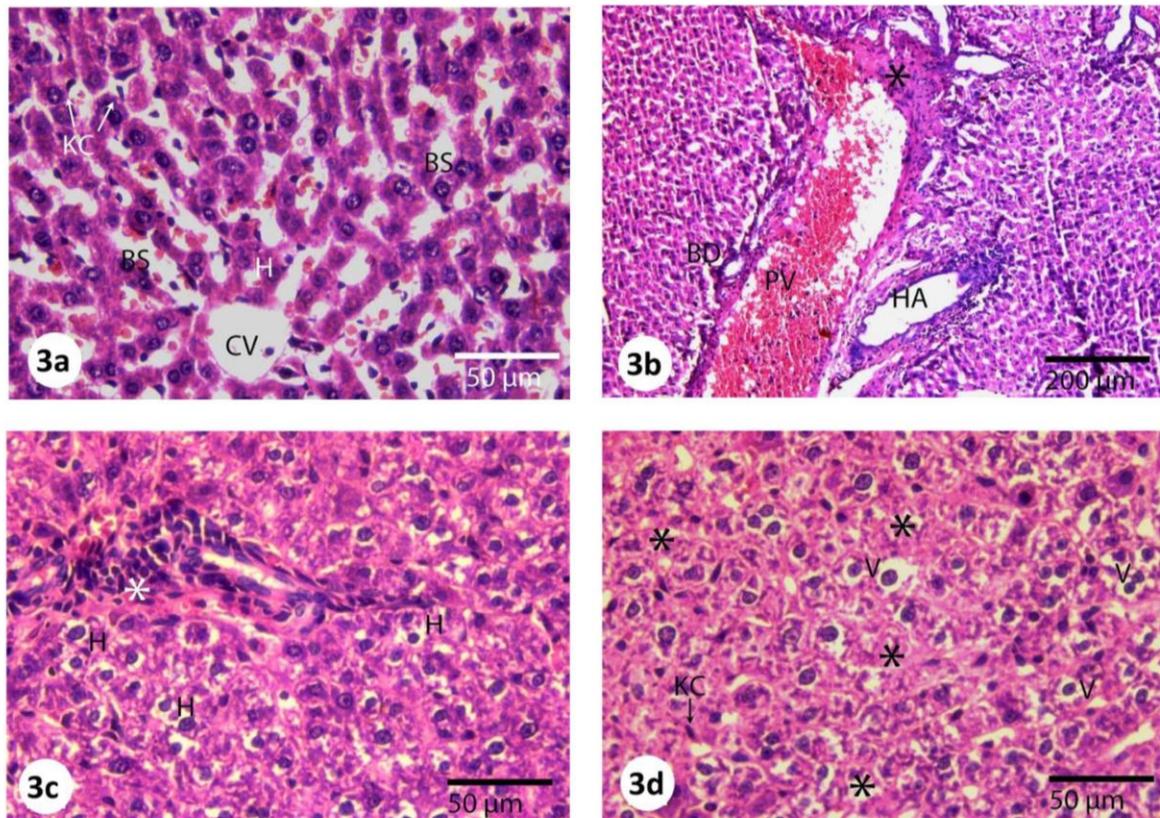
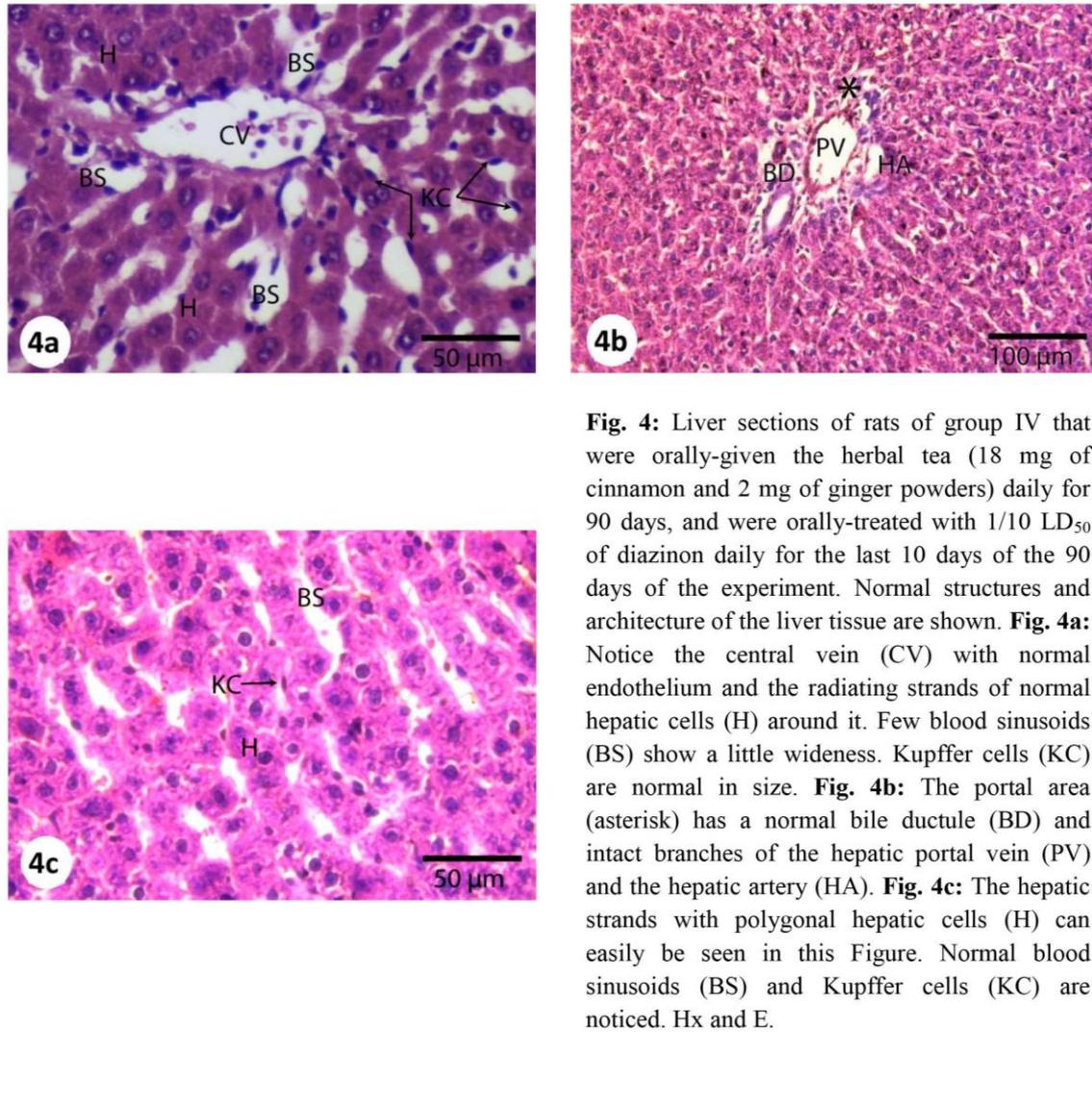


Fig. 3: Liver sections of rats of group III that were orally-treated with 1/10 LD₅₀ of diazinon daily for the last 10 days of the 90 days of the experiment. **Fig. 3a:** Notice the widening of the blood sinusoids (BS). The endothelium of the central vein (CV) and some of the liver cells around it display degeneration. Hypertrophied Kupffer cells (KC) are also seen. **Fig. 3b:** The portal triad shows deformation and loss of the hepatic architecture. Notice the branch of the hepatic portal vein (PV) appears swollen and massively congested with blood. The cells of the limiting plate in the portal area are completely deformed, and in some areas (asterisk) become very thick. Notice the branch of the hepatic artery (HA) that exhibits marked swelling. **Fig. 3c:** Many inflammatory cells are infiltrated (asterisk) among the degenerated hepatocytes (H). **Fig. 3d:** The majority of the hepatic cells (H) show vacuolation, shrunken cytoplasm, pleomorphic nuclei, and others have shrunken nuclei. Some necrotic areas (asterisk) are also noticed. Hx and E.



Electron microscopic results**Group I (control group) and group II (tea group) – (Figs. 5a-b and 6a-b):**

Electron microscopic examination of the liver sections of the control rats of both groups I and II showed normal ultrastructure of the liver tissue. The different ultrathin sections of the liver of these control rats showed the polygonal boundaries of the hepatocytes. The cytoplasm of the hepatocytes embodies many glycogen granules. The rough endoplasmic reticulum exhibits normal cisternae. The mitochondria are spherical to oval in shape with their characteristic transverse cristae. The Golgi apparatus appears as a few closely packed flat saccules that lie near the bile canaliculi. The nuclei of the liver cells are prominent and spherical in shape. The nuclear pores are easily distinguished along the doubled membranous nuclear envelope. Most of the nuclei have one nucleolus or occasionally two nucleoli. The nucleoli are spherical and consist of fine dark fibrils and dense granules. The location of the nucleoli varies inside the nuclei; central, eccentric or peripheral. The nuclei also present prominent granular clumps of chromatin filaments. The Kupffer cells are spindle-shaped and possess many cytoplasmic processes. They have clear vacuoles and occasionally, dark bodies according to their activity. Their nuclei are elongated and have electron-dense chromatin. Normal bile canaliculi were clearly seen. They have many intact microvilli that project from the plasmalemmae of the adjacent hepatocytes.

Group III (diazinon group) – (Figs. 7a-d):

The ultrathin sections of the liver of rats treated daily with 1/10 of the LD₅₀ of diazinon in corn oil, for the last 10 days of the 90 days of the experiment revealed many cytopathological changes. The boundaries of the hepatocytes displayed marked distortion. The cytoplasm of the liver cells exhibited

shrinkage and possessed a few lipid droplets. The endoplasmic reticulum became swollen and deformed. The mitochondria of the hepatocytes showed different degrees of degeneration and became less in number than those of the control liver cells. Golgi apparatus was not seen due to its degeneration. Many liver cells had pyknotic nuclei. Other cells were necrotic and appeared as large residual bodies which were expelled into the lumens of the blood sinusoids. Many squeezed red blood cells surrounded by perisinusoidal processes were present inside the markedly-swollen blood sinusoids. The bile canaliculi exhibited deformation and their microvilli became very few and appeared as ghosts. The phagocytic inflammatory cells, attacked the tissue; they had indented nuclei and engulfed remnant parts of the degenerated liver cells.

Group IV (tea and diazinon group) – (Figs. 8a-b)

The ultrathin sections examined by using the electron microscope showed that the membranes of the hepatocytes were completely intact and the adjacent hepatic cells were easily distinguished from each other. The hepatocytes had normal cytoplasm. Their cytoplasmic organelles such as mitochondria, Golgi apparatus, ribosomes and endoplasmic reticulum were completely normal in their appearance. The nuclei of the liver cells were intact and had normal structure. All other features of the control liver examined by using the electron microscope were clearly seen in the electron micrographs of the ultrathin liver sections of the rats of group IV. These ultrastructure findings confirmed the hepatoprotective effect – revealed by the histological examination using the light microscope – of the cinnamon and ginger tea against the hepatotoxicity induced in rats by the insecticide diazinon.

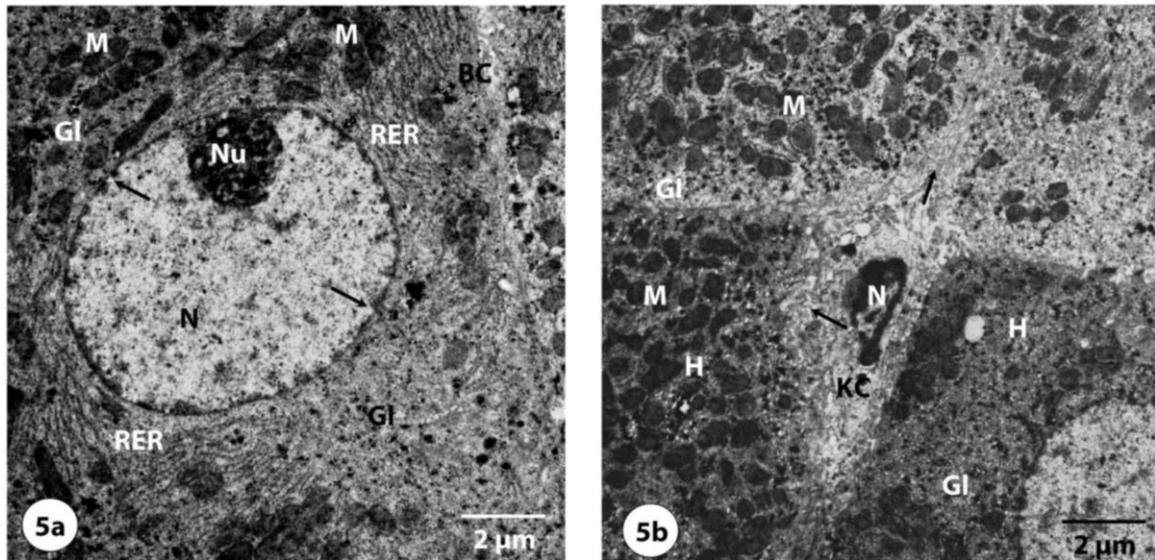


Fig. 5: Electron micrographs of ultrathin sections of liver of untreated control rats of group I showing in **Fig. 5a** a hepatocyte that has a normal cytoplasm and a spherical nucleus (N). Mitochondria (M), rough endoplasmic reticulum (RER), and glycogen granules (GL) are noticed. The nucleus has a spherical nucleolus (Nu) and nuclear pores (arrows) in the double-membrane nuclear envelope. At the upper right corner, a bile canaliculus (BC) is shown. **Fig. 5b:** Notice Kupffer cells (KC) with their elongated and darkly-stained nuclei (N) and many cytoplasmic processes (arrows).

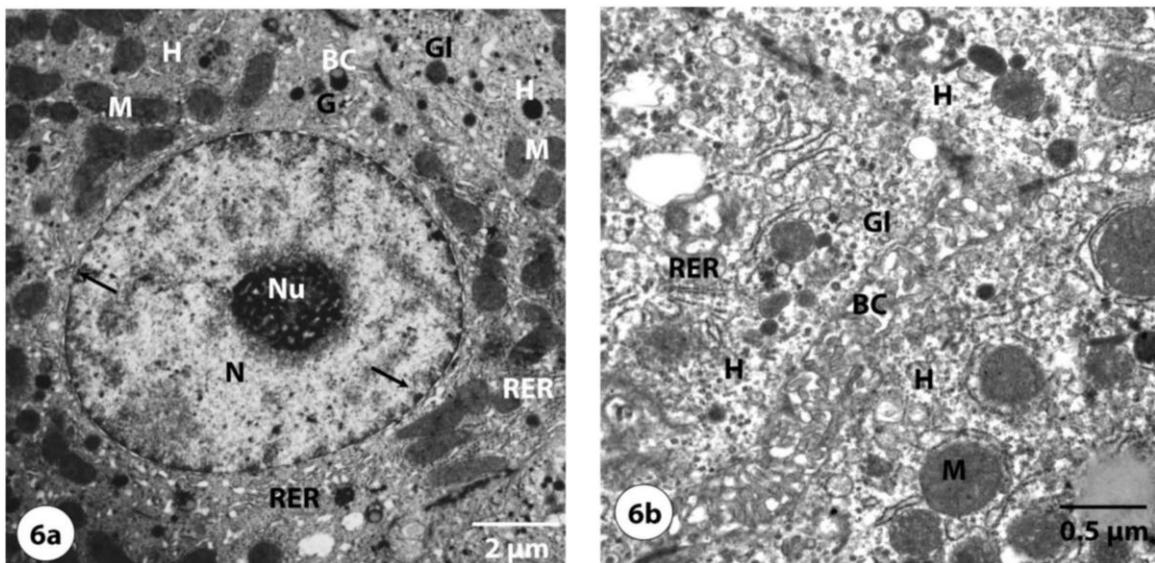


Fig. 6: Electron micrographs of ultrathin sections of liver of rats of group II that were orally-given the herbal tea (18 mg of cinnamon and 2 mg of ginger powders) daily for 90 days. **Fig. 6a** shows normal structures of the hepatocytes (H); nucleus (N) and nucleolus (Nu), rough endoplasmic reticulum (RER), mitochondria (M), Golgi apparatus (G) and glycogen granules (GL). **Fig. 6b:** A magnified micrograph showing cytoplasmic parts of three hepatocytes (H) that send their projections as many microvilli into the lumen of a bile canaliculus (BC).

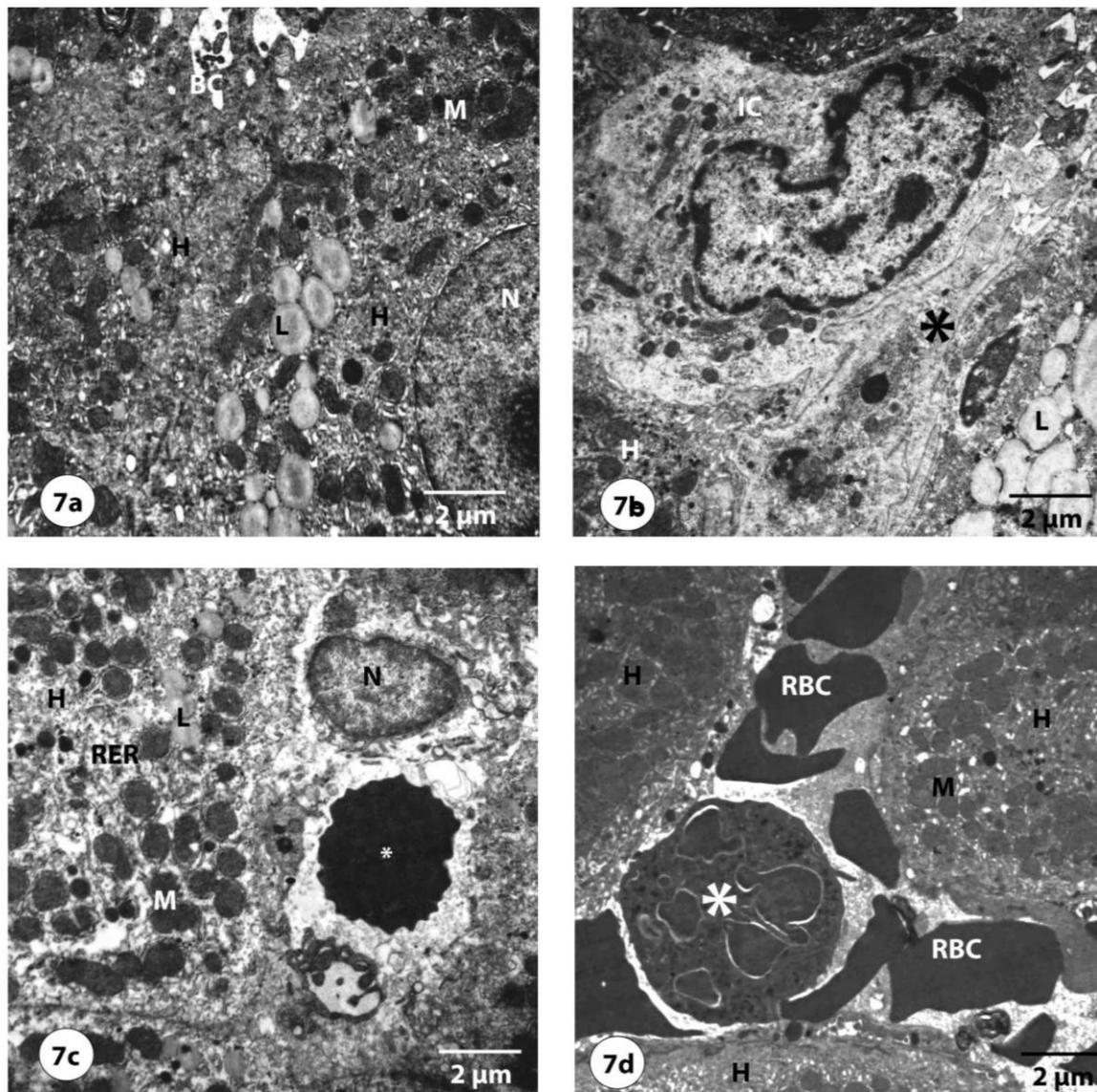


Fig. 7: Electron micrographs of ultrathin sections of liver of rats of group III that are orally-treated with 1/10 LD₅₀ of diazinon daily for the last 10 days of the 90 days of the experiment. In **Fig. 7a** cytoplasmic parts of two hepatocytes (H) are hardly differentiated from each other due to distortion of the boundaries between them. Notice the presence of a few lipid droplets (L) within the cytoplasm. The mitochondria (M) are few and have different degrees of damage. There are two deformed bile canaliculi (BC) at the upper border of the micrograph. **Fig. 7b:** An inflammatory cell (IC) engulfing a degenerated remnant (asterisk) of the liver tissue. **Fig. 7c:** A necrotic hepatocyte appears having a shrunken cytoplasm and a pyknotic nucleus (N) above a residual degenerated body (asterisk). The rough endoplasmic reticulum (RER) shows swelling and deformation. Many mitochondria (M) are damaged. **Fig. 7d:** Notice presence of a swollen blood sinusoid that has a large residual body (asterisk) and many squeezed red blood cells (RBC) surrounded by perisinusoidal processes.

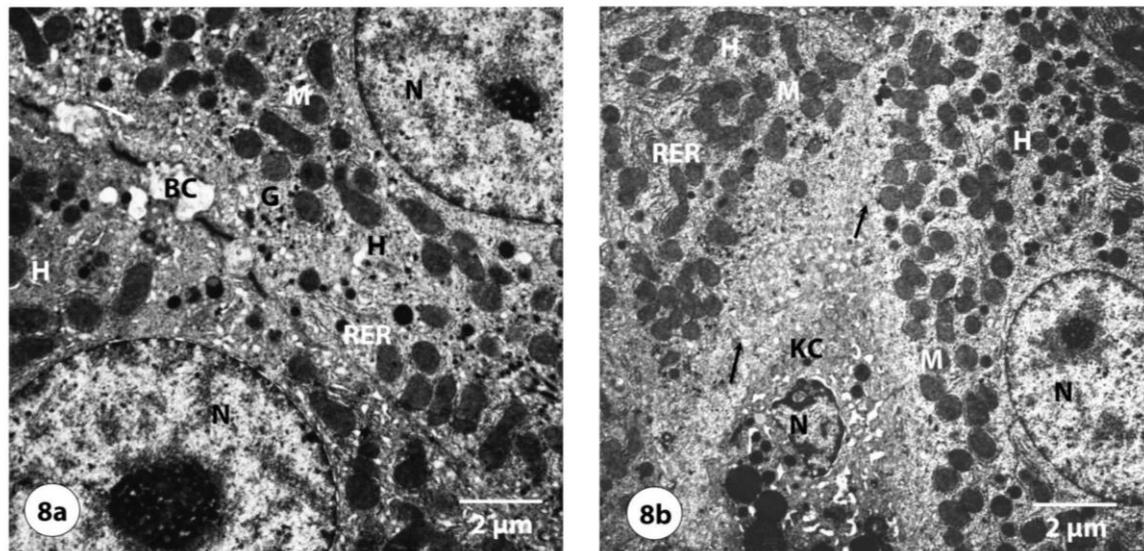


Fig. 8: Electron micrographs of ultrathin sections of liver of rats of group IV that were orally-given the herbal tea (18 mg of cinnamon and 2 mg of ginger powders) daily for 90 days, and were orally-treated with 1/10 LD₅₀ of diazinon daily for the last 10 days of the 90 days of the experiment. **Fig. 8a:** Parts of the cytoplasm and the nuclei (N) of two adjacent hepatocytes (H) are shown. Normal organelles are present: mitochondria (M), rough endoplasmic reticulum (RER), and Golgi apparatus (G). The bile canaliculi (BC) are seen between the two hepatocytes. **Fig. 8b:** Among the hepatocytes (H), there are Kupffer cells (KC) with their elongated and darkly-stained nucleus (N) and many cytoplasmic processes (arrows).

DISCUSSION

The present study revealed the histopathological and ultrastructural alterations insulted the liver of rats due to induced hepatotoxicity by the organophosphorous insecticide diazinon. The current study showed that diazinon caused marked hepatotoxic effect in the rats treated with low daily-doses (1/10 LD₅₀) for the last 10 days of the 90 days of the experiment. Almost, similar findings of diazinon-induced hepatotoxicity features were revealed in previous studies carried out by many other authors. Necrotic areas were observed in liver of rats treated with a single dose of diazinon; 200mg/kg body weight³⁶. Liver of diazinon-exposed mice showed vacuolization of the hepatocytes, infiltration of inflammatory cells, congestion of the central veins, and an increase in the number of Kupffer cells³⁷. Another study showed that diazinon caused widening of the blood sinusoids, congestion of the central veins, and deformation of the portal triad, in addition to induction of haemorrhage, necrosis, and

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lymphoid infiltration in rats' liver³⁸. In electron microscopic investigations of hepatocytes of diazinon-treated Wistar rats, the mitochondria exhibited swelling and breaking up of their cristae³⁹. Also, diazinon induced histopathological changes in liver tissue of rats by oxidative stress mechanisms; of these changes were disruption of the radial alignment of the hepatocytes around the central veins, cytoplasmic vacuolization of the hepatocytes, inflammation signs around the central veins and the portal areas⁴⁰. Other data showed that inhalation of diazinon by pregnant mice increased the apoptosis of the hepatocytes of the embryo⁴¹. Other studies suggested that diazinon may induce hepatotoxicity through oxidative stress^{42, 43}, apoptosis, and metabolic disorders in liver⁴³. It was reported that the acetyl cholinesterase did not play a role in the liver damage induced by diazinon^{44, 45}.

As far as the present author is aware, no earlier study on the impact of the present herbal mixture on the induced hepatotoxicity

by diazinon. And taking into consideration the economical conditions of the Egyptian farmers and cattlemen who are apt to the dangers of the organophosphorous insecticides such as diazinon, it was very important to seek ameliorative natural products that are safer and cheaper than the common medication or drugs against the adverse effects of such pollutants. On applying the present plan in this study, the results revealed that rats given the cinnamon and ginger tea for 90 consecutive days had no histopathological or cytopathological signs of hepatotoxicity.

Many previous studies proved that the cinnamon⁴⁶⁻⁴⁸ or ginger⁴⁹⁻⁵³ extracts have antioxidant activities and can combat hepatotoxicity induced by some insecticides. Cinnamon and ginger extracts induced antioxidant effects in obese diabetic rats⁵⁴. Similar studies were carried out on mixtures of herbal products such as combination of barley flour and cinnamon that showed antioxidant activity⁵⁵. Other studies revealed that a mixture of cinnamon, ginger, and clove extracts improved testicular histopathology, testosterone levels and sperm quality of diabetic rats due to its antioxidant constituents⁵⁶. Therefore, the current results agree with previously-mentioned studies supposing that the ameliorative role of the aqueous powders mixture of cinnamon and ginger against the hepatotoxicity induced by diazinon is due to their antioxidant constituents.

This hepatoprotective role of the mixture needs more research for the biochemical and enzymes assessment to compare the degree of the amelioration role between the mixture of cinnamon and ginger tea and the single compounds – cinnamon tea or ginger tea, solely – under the same conditions and same preparation manner.

Conclusion and Recommendation

The above-mentioned results suggest that the cinnamon and ginger tea apparently counteracted and suppressed the development of the signs of the hepatotoxicity induced by diazinon in rats. In the light of the present findings, it is advised to consider drinking the

cinnamon and ginger tea as a manner of avoiding the dangerous effects of environmental pollutants such as the organophosphorous insecticides.

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