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ORIGINAL ARTICLES

Biochemical and Histopathological Effects of Systemic Pesticides on Some Functional Organs of Male Albino Rats

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ABSTRACT

This research was carried out to throw the light on the biochemical alterations including liver function parameters; serum aminotransferase (AST and ALT) and alkaline phosphates (ALP) activities, and kidney function parameters; serum urea and creatinine levels, in male albino rats as affected by the daily orally administration of a single dose equivalent to either 1/20 or 1/10 from $LD_{50\%}$ of dimethoate, carbofuran and carbendazim pesticides individually for 30 days. The present study also included the histopathological effects of the former daily orally intake from the tested systemic pesticides on liver and kidney tissues of albino rats. The current results showed a significant decrease in body weights of rats treated with high dose (1/10 $LD_{50\%}$) of tested pesticides. While, rats treated with low dose (1/20 $LD_{50\%}$) of these pesticides showed non-significant alterations in the body weights when compared with the control group. On the other hand there was a significant increase in the relative liver and kidney weights of rats-treated with the high dose (1/10 $LD_{50\%}$) all tested pesticides. While, there was no significant alteration in the relative liver and kidney weights in those treated with low dose (1/20 $LD_{50\%}$) of tested pesticides. Biochemical analysis results illustrated that there was an exceptional rise at the end of experiment in liver functional AST, ALT and ALP enzymes activity and a significant increase ($P < 0.05$) in urea and creatinine levels in serum of rats, especially with the high dose treatment, compared to the control untreated rats group. Histopathological examination exhibited that there were histological changes observed in the liver of rats received high dose of tested pesticides; including pyknotic nuclei, focal necrosis with inflammatory infiltration, vacuolation and blood congestion, while rats treated with low doses of tested pesticides showed that normal hepatocytes as compared with the control group. Histopathological examination of kidneys revealed that there was some blood congestion in between tubules and small area of hemorrhage in the interstitium in rats treated with high doses of pesticides. But, rats received low doses of tested pesticides showed normal structure of renal corpuseles and tubules comparing with control animals.

Key words: Pollution, Systemic pesticides, Biochemical alteration, Liver functions, Kidney function, Histopathological effects.

Introduction

The environmental pollution is one of the most serious problems that faces mankind in this century. There are many types of pollutants that interfere with our-life both directly and indirectly. Furthermore, potential future hazards to human health and wildlife can be created by residues from some long-lived pesticides, that may build up in the food chain and cause widespread contamination of the environment (El-Sebae, 1993; Zaahkouk *et al*., 2000). There has been growing concern about the indiscriminate and excessive use of pesticides and the consequent environmental pollution and adverse health effects on man. In recent years, the hazards of using these chemicals have been accentuated by the sharp rise in their use in agriculture and industry and by house holders and governments (Hagar and Fahmy, 2002; Heudorf *et al*., 2006 and Sayim, 2007).

Pesticides are biological active chemicals used in agriculture to destroy or control weeds, insects, fungi and other pests. When pesticides are applied improperly, the resulting residues in the produce can pose significant health risks to consumers, who are increasingly aware of the potential for contamination of food and drinking water (Hotchkiss, 1992 and Chavarri *et al.,* 2004). Some of these pesticides are regarded as systemic when it penetrates into the treated plants through the vascular system or leaf cuticle and kills piercing and sucking pest insects and pests which feed on sap or cell content (Abou-Arab., 1999; Tasei *et al*., 2003). Dimethoate, carbofuran and carbendazim are the widely used systemic pesticides in agriculture against a wide range of insects, mites and fungal diseases of fruits, vegetables, or namental plants and field crops as both systemic and contact pesticides and are also used indoor to control houseflies (Meister, 1992; Farag *et al*., 2006; Brkic *et al*.,

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2008 and Muthuvivegandave *et al*., 2011). The irrational and excessive use of these pesticides play a crucial role in the occurrence of many disease affecting plants, animals and human (Zaahkouk *et al*., 2000 and Al-Haj *et al*., 2005).

Poisoning of the former systemic pesticides is a well known toxicological problem in developing countries, but well still has, even in industrialized ones, a high mortality rate and a frequent invalidating outcome (Lifshitz *et al*., 1994). Systemic pesticides have been implicated in various disorders and diseases including cancer, adverse reproductive outcomes, peripheral neuropathies, disorders, impaired immune functions and allergic sensitization reactions, particularly of the skin, cumulative inhibition of cholinesterase activity as a result of long-term low doses of exposure to these pesticides (WHO/UNEP, 1990; Hagar and Fahmy, 2002 and Khogali *et al*., 2005). It has peen reported the toxicity of systemic pesticides results in deleterious effects on many organs and systems in human and other mammals particularly the nervous system (Nagymajtenyi *et al*., 1998 and Hagar and Fahmy, 2002), immune system (Aly and El-Genl, 2000), reproductive system and sexual hormones (Rawling *et al*., 1998 and Muthuvivegandave *et al*., 2011), liver (Gomes *et al*., 1999 and Selmanoglu *et al*., 2001), Kidney (Khogali *et al*., 2005 and Brkic *et al*., 2008), pancreas (Hagar and Fahmy, 2002), brain (Hunt and Hooper, 1993 and Khogali *et al*., 2005). Some biochemical alterations took place in human and other experimental animals due to the systemic pesticide toxicity were reported and included inhibition of both mammals brain and plasma cholinesterase activity (Hunt and Hooper, 1993; Zaahkouk *et al*., 2000 and Berny *et al*, 2007), and mammals erythrocytes and plasma acetyl cholinesterase enzyme activity of liver functional enzymes including serum aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP) and lactate dehydrogenase (Begum, 2004; Sayim, 2007 and Salih, 2010), and the increase in serum urea, uric acid and creatinine (Selmanoglu *et al*., 2001 and Soufy *et al*., 2007). It has been reported that there are some histopathological alterations in liver, kidney and testis of human and other mammals due to the exposure for some systemic pesticides including mononuclear cell infiltration, congestion, an enlargement of the veins and sinusoids, necrotic changes, an increase in the number of Kupffer cells, cytoplasmic vacuolization, degeneration in nuclei in the liver, hydropic degeneration and hepatocellular damage in the liver, damage in thyroid, parathyroid and adrenal glands (Selmanoglu *et al*., 2001; Barlas *et al*., 2002; Sayim, 2007 and Muthuviveganandave *et al*., 2011).

So far, few efforts have been made and limited published information are available on the biochemical and histopathological effects of systemic pesticides on the functional organs in mammals. Therefore, this research was performed to throw the light on the biochemical alterations including liver function parameters; serum aminotransferase (AST and ALT) and alkaline phosphates (ALP) activities, and kidney function parameters; serum urea and creatinine levels, in male albino rats as affected by the daily orally administration of a single dose equivalent to either 1/20 or 1/10 from $LD_{50\%}$ of dimethoate, carbofuran and carbendazim pesticides individually for 30 days. The present study also included the histopathological effects of the former daily orally intake from the tested systemic pesticides on liver and kidney tissues of albino rats.

Materials and Methods

Materials:

Dimethoate; Cheminova 40% E.C.; [O, O-dimethyl S-(2- (methylamino) -2- oxoethyl) phosphoro dithioate. The acute oral $LD_{50\%}$ for rats 291-325 mg active ingredient /kg body weight. Carbofuran; Furadan 10% G.R.;[2, 3-dihydro -2, 2-dimethyl-7-benzofuranyl methyl carbamate]. The acute oral LD_{50} for rats 8 mg active ingredient /kg. body weight. Carbendazim; Bendazine 50% W.P.; [1-methyl 1-*H*-benzimidazol-2-yl-carbamate].The acute oral LD_{50} for rats 6400 mg active ingredient /kg body weight. Chemical and kits used in biological analysis and histopathological examination where in analytical great, and purchased from El-Gamhouria Trading Chemicals and Drugs Company, Egypt.

Methods:

Experimental Animals:

Adult male albino rats, weighting 120 - 130 gm were purchased from the Biological Products & Vaccines Holding Company, Helwan Farm, were used in this study. The animals were housed in groups of 7 in stainless steel community cages at 22 ± 2 ^{o}C and 65 ± 5% R.H. with a 12 hr. light/dark cycle and allowed to acclimatize for a period of 15 days prior to experimental use. The animals were maintained on commercial standard pellet diet purchased from Egyptian Company of Oils and Soaps, Composed of 21% protein, 2% fat, 12.8% Can starch, 4.2% Salt and vitamins mixture and vitamins, and water adlibitum.

J. Appl. Sci. Res., 8(11): 5459-5469, 2012

Experiment Design:

Forty-nine rats were used and classified into seven groups, each group had seven rats. Group (1) was served as control animals, they received 1 ml. of corn oil; group (2) Rats given daily an oral low dose equivalent to (1/20 LD_{50}; 20 mg/kg b.w.) of dimethoate for 30 days; group (3) Rats given daily an oral high dose equivalent to (1/10 LD_{50}; 40 mg/kg b.w.) of dimethoate for 30 days; group (4) Rats given daily an oral low dose equivalent to (1/20 LD_{50}; 0.4 mg/kg b.w.) of carbofuran for 30 days; group (5) Rats given daily an oral high dose equivalent to (1/10 LD_{50}; 0.8 mg/kg b.w.) of carbofuran for 30 days; group (6) Rats given daily an oral low dose equivalent to (1/20 LD_{50}; 320 mg/kg b.w.) of carbendazim for 30 days; group (7) Rats given daily an oral high dose equivalent to (1/10 LD_{50}; 640 mg/kg b.w.) of carbendazim for 30 days. At the end of experimental period, blood samples were collected from the retro-orbital sinus plexus from all animals after being fasted for 12 hours for different biochemical analysis. Blood samples were left to clot and centrifuged at 5000 rpm at 4 ^{o}C for 10 min to separate the serum.

Histopathological examination carried out according to (Sarkar *et al*., 2005), liver and kidneys were dissected out and fixed instantaneously in 10% formal saline for 24 hours. The specimens were washed in tap water, dehydrated in ascending grades of ethanol, cleared in xylene, embedded in paraffin wax (melting point of 50-56 ^{o} C). Paraffin sections were cut at 6µm thicknesses using a rotary microtome (Model MR 60, Russian); the sections were stained with Harris haematoxylin and eosine. Observation were made using a light microscope (Zeiss Axiophot, Germany) and photographs were taken with an automatic photomicrographic system.

Biochemical analysis were performed: serum aspartate transaminase (AST) and serum alanine transaminase (ALT) activity were carried out according to the colourimeteric method of Schmidt and Schmidt (1963), serum alkaline phosphatase (ALP) activity was determined according to calorimetric method of Belfield and Goldberg (1971). Blood urea was estimated by the enzymatic method of Patten and Crouch (1977) and Serum creatinine was determined according to the method described by Faulkner and King (1976). Liver index was calculated (liver weight/body weight x 100%) and Kidney index was calculated (kidney weight/ body weight x 100%) by Ping *et al*. (2006).

Statistical Analysis:

All obtained results are expressed as mean ± standard error. The statistical comparison between the initial group at the beginning of experiment. Control untreated group and systemic pesticides-treated groups was performed by using a one-way analysis of variance (ANOVA) followed by Duncan's test according to the procedure of Armitage (1971) using SPSS version 11 computer program.

Results:

1. Effects of The Oral Administration of Tested Systemic Pesticides on Body Weight and Relative Organs Weights of Tested Male Albino Rats:

The effects of the oral administration of either the low dose equivalent (1/20 $LD_{50\%}$) and high dose (1/10 $LD_{50\%}$) of dimethoate, carbofuran and carbendazim pesticides given daily individually for 30 successive days on body weight of male albino rats are presented in

Table (1). The present results (Table 1) evident that animals in all treatment group showed an increase in body weight over the duration of experimental period which could be attributed to the normal growth phase and the hyperlipidemic diets. But, animals received the high dose of dimethoate, carbofuran and carbendazim showed that significant decrease in body weight as compared with control group and groups treated with low dose of tested pesticides. These decrease in body weights started after 14 days and continued to the end of experiment.

Table 1: Alterations in body weight of male albino rats throughout thirty days of the oral administration of tested pesticides:

Treatment	Dose	Body weight (M±SE)▪		
		At the beginning of experiment	After 15 days of treatment	At the end of experiment (30 days)
Control	-	132 ± 0.70	172 ± 0.83	205 ± 2.61
Dimethoate	Low	131 ± 0.72	168 ± 0.63*	194 ± 1.87*
	High	129 ± 0.74	155 ± 0.54*♦	184 ± 2.07*♦
Carbofuran	Low	130 ± 0.70	164 ± 0.83*	196 ± 2.60*
	High	128 ± 0.83	152 ± 0.57*♦	179 ± 1.51*♦
Carbendazim	Low	130 ± 0.80	160 ± 1.14*	197 ± 2.54*
	High	128 ± 0.82	150 ± 0.63*♦	185 ± 2.91*♦

Low dose: equivalent 1/20 from $LD_{50\%}$; High dose: equivalent 1/10 from $LD_{50\%}$; ▪ Mean ± standard error for body weight.;
** Significantly different as compared with the control group ($P < 0.05$);♦ Significantly different as compared with groups treated with low doses ($P < 0.05$.)*

From the obtained data (Table. 2), it could be observed that there was a significant increase ($P < 0.05$) in the relative liver weights of treated rats with high dose of tested pesticides compared with control group and low dose groups. While, the experimental animals treated with low dose of either dimethoate, carbofuran and carbendazim showed non significant increase ($P \leq 0.05$) in the relative liver and kidney weights comparing with the control animals.

Table 2: Relative liver and kidneys weights of male albino rats after thirty days of oral administration of tested pesticides:

Description	Tested parameter at the end of experiment period (30 days) ▪						
	Control group	Dimethoate-treated group		Carbofuran-treated group		Carbendazim-treated group	
		Low dose	High dose	Low dose	High dose	Low dose	High dose
Body weight (g)	205 ± 2.61	194 * ± 1.87	184 *♦ ± 2.07	196 * ± 2.60	179 *♦ ± 1.51	197 * ± 2.54	185 *♦ ± 2.91
Liver weight (g)	5.87 ± 0.35	6.68 * ± 0.46	7.88 *♦ ± 0.38	6.83 * ± 0.32	7.91 *♦ ± 0.12	6.89 * ± 0.37	7.93 *♦ ± 0.22
Liver index (%)	2.863	3.443	4.282	3.484	4.418	3.497	4.286
Kidney weight (g)	1.28 ±0.07	1.30 ± 0.04	1.44 * ± 0.04	1.33 ± 0.07	1.42 * ± 0.11	1.31 ± 0.04	1.40 * ± 0.05
Kidney index (%)	0.624	0.670	0.782	0.678	0.793	0.664	0.756

*Low dose: equivalent 1/20 from $LD_{50\%}$; High dose: equivalent 1/10 from $LD_{50\%}$.; ▪ Mean±Standard error; the means within the same row having different superscript are significantly different ($P < 0.05$).; * Significant different at $P < 0.05$ as compared with control group.; ♦ *Significant different at $P < 0.05$ as compared with low dose group.*

2. Effect of Oral Administration of Tested Systemic Pesticides on Liver and Kidney Functions of Male Albino Rats:

Liver and kidney functions parameter in the serum of male albino rats as affected by the oral administration of tested systemic pesticides; dimethoate, carbofuran and carbendazim, individually for thirty days were investigated and the obtained results are recorded as in Table (2). From the current data (Table 2), it could be noticed that there was an exceptional rise in tested liver functional enzymes; aspartate amino transferase (AST), alanine amino transferase (ALT) and alkaline phosphatase (ALP) activity in serum of experimental animals treated with all tested individual systemic pesticides. The elevation rate in the former tested functional liver enzymes was increased significantly ($P \leq 0.05$) with increasing the oral intake dose and extending the dietary intake period of all tested pesticides. Whereas; the AST, ALT and ALP activities were 21.3, 30.1 and 181.8 IU/L in serum of the initial untreated group, while the former corresponding liver functional enzymes activities in the control untreated rats group at the end of experiment period (after 30 days) were 22.5, 29.8 and 179.3; respectively. On the other hand, serum AST activity value in tested pesticides-treated rats groups at the end of experiment were 139.2, 156.7 and 137.9 IU/L for rats groups treated with low dose (1/20 $LD_{50\%}$) from dimethoate, carbofuran and carbendazim, versus 201.6, 221.3 and 184.6 IU/L for those treated with high dose (1/10 $LD_{50\%}$) from the former corresponding systemic pesticides; respectively. Serum ALT activity values in rats' groups treated with low dose of dimethoate, carbofuran and carbendazim were 110.4, 123.1 and 94.6 IU/L, against 159.9, 190.2 and 140.7 IU/L for those received the high dose of the corresponding pesticides at the end of experiment (after 30 days); respectively. In addition, serum ALP activity values in rats groups treated with low dose of either dimethoate, carbofuran or carbendazim at the end of experiment were 141.6, 438.9 and 409.1IU/L, while the corresponding values for those treated with high dose of the former pesticides were 577.3, 635.4 and 515.6, respectively.

With regards kidney functions as shown in Table (3), there was significant increase ($P \leq 0.05$) in kidney functions' parameters; serum urea and creatinine, of treated experimental animals' group with all individual systemic pesticides for 30 days as pesticide dose and dietary period were increased. Serum urea and creatinine levels were 27.4 and 0.62 mg/dl in initial un treated rats group, while the levels of corresponding kidney function parameters were 26.9 and 0.64 mg/dl in serum the control untreated rats group; at the end of experiment. The obtained data (Table 3) also illustrated that serum urea values after 30 days of the orally given of low dose from dimethoate, carbofuran and carbendazim were 49.3, 51.6 and 48.4 mg/dl, versus 68.7, 70.1 and 63.9 mg/dl for those treated with high dose of former systemic pesticides; respectively. furthermore, serum creatinine levels in the rats' groups orally treated with low dose of the former pesticides for 30 days were 1.29, 1.42 and 1.29 mg/dl, against 1.86, 2.05 and 1.83 mg/dl for those treated with high dose of the former pesticides for the same period; respectively.

Table 3: Effect of the oral administration of tested systemic pesticides on liver and kidney functions' parameters:-

Function Parameter	Tested function parameter value (M±SE)▪ At the end of experiment period (30 days)								Normal value▲
	Initial group	Control group	Dimethoate		Carbofuran		Carbendazim		
			Low dose	High dose	Low dose	High dose	Low dose	High dose	
AST (IU/L)	21.3[a] ± 1.07	22.5[a] ± 1.29	139.2[b] ± 4.89	201.6[e] ± 4.89	156.7[c] ± 4.31	221.3[f] ± 5.70	137.9[b] ± 4.13	184.6[d] ± 5.66	Up to 37
ALT (IU/L)	30.1[a] ± 1.19	29.8[a] ± 1.05	110.4[c] ± 3.79	159.9[f] ± 4.46	123.1[d] ± 3.72	190.2[g] ±4.13	94.6[b] ± 3.79	140.7[e] ± 5.13	Up to 41
ALP (IU/L)	181.8[a] ± 4.76	179.3[a] ± 5.21	441.6[c] ± 12.87	577.3 ± 13.09	438.9[c] ± 11.63	635.4[e] ± 12.89	409.1[b] ± 12.07	515.6[d] ± 13.81	68 - 400
Urea (mg/dl)	27.4[a] ± 0.39	26.90[a] ± 0.46	49.3[bc] ± 0.67	68.7[e] ± 1.89	51.6[c] ± 1.13	70.1[e] ± 1.26	48.4[b] ± 0.53	63.9[d] ± 1.47	15 - 45
Creatinine (mg/dl)	0.62[a] ±0.023	0.64[a] ± 0.031	1.29[b] ± 0.026	1.86[d] ± 0.041	1.42[c] ± 0.039	2.05[e] ± 0.050	1.29[b] ± 0.027	1.83[d] ± 0.071	0.5 – 1.5

▪ *Mean ± Standard error for function parameter; the means within the same row having different superscripts are significantly varied;* ▲ *Normal value of each function parameter in the serum of healthy experimental male albino rats reported by Murray et al. (1991) and Kaneko et al. (1997).; Low dose: equivalent 1/20 $LD_{50\%}$; High dose: equivalent 1/10 $LD_{50\%}$.*

3. Histopathological Alterations in Some Functional Organs of Male Albino Rats as Affected by the Oral Administration of Tested Systemic Pesticides for 30 Days:

a. Liver Histopathological Alterations:

The histopathological examination of the liver sections in the control untreated rats group showed a normal histological picture. The central vein lies at the centre of the lobule surrounded by the hepatocytes with strongly eosinophilic granulated cytoplasm, and distinic nuclei. In addition, between the strands of hepatocytes the hepatic sinusoids are exhibited as shown in Figure (1a). The liver of rats treated with low dose of dimethoate showed a normal structure of a hepatic lobule, and the activated Kupffer cells were noticed as given in Figure (1b). The picture of the liver sections of rats treated with high dose of dimethoate showed that there were focal necrosis with inflammatory infiltration and was also notice hepatic sinusoids congestion as evident in Figure (1c). Also, liver section of some rats in this group treated with high dose of dimethoate showed vacuoles in the hepatocytes and congested hepatic sinusoids (Figure 1c & Figure 1d). With regards the liver sections of rats group treated with low dose (equivalent to 1/20 $LD_{50\%}$) of carbofuran as shown in Figure (1e), there were normal hepatocytes and dilated hepatic sinusoids congestion (arrow). While, the histopathological examination picture of liver sections for the rats group received an oral high dose (equivalent to 1/10 $LD_{50\%}$) of carbofuran (Figures 1f & 1g) exhibited a large vacuoles in the hepatocytes and the hepatic sinusoids congestion (Figure 1f). Also, some liver sections for some rats of this group showed that there was disturbance of the hepatic lobules and also microvesicle and pyknotic nuclei were observed as evident in Figure (1g). Concerning the rats group treated with the low dose (equivalent to 1/20 $LD_{50\%}$) of carbendazim, the histopathological examination photomicrographs for liver sections of this group (Figure 1h) showed a normal hepatocytes. While, the corresponding photomicrographs for liver sections of experimental animals group given an oral high dose (equivalent to 1/10 $LD_{50\%}$) of carbendazim (Figures 1i & 1j) evident that there were a congested blood vessel in the portal area (arrow) and few inflammatory infiltration (arrow) as shown in Figure (1i). Also, liver section of some rats in this group treated with the high dose of carbendazim exhibited that there were disturbance of the hepatic lobules and also the microvesicle and pyknotic nuclei were noticed as illustrated in Figure (1j).

b. Kidney Histopathological Alterations:

Histopathological examination photomicrographs of the kidney sections in the control group showed a renal corpuscle and renal tubules, proximal convoluted tubules and distal convoluted tubules. Also, the lomerulus, urinary space and Bowman's capsule were noticed as shown in Figure (2a).

From the histopathological examination photomicrographs for kidney sections of rats group given an oral low dose equivalent to 1/20 $LD_{50\%}$ of either dimethoate (Fig. 2b), carbofuran (Fig. 2d) and carbendazim (Fig. 2f), it could be observed a renal corpuscle, renal tubules, proximal convoluted tubules (PCT) and distal convoluted tubules (DCT). In addition that the glomerulus (G), urinary space (US) and Bowman's capsule were observed as shown in Fig. (2b), Fig. (2d) and Fig. (2f).

Photomicrographs for kidney sections of rats given a high dose equivalent to 1/10 $LD_{50\%}$ of either dimethoate (Fig. 2c), carbofuran (Fig. 2e) or carbendazim (Fig. 2g) exhibited that there were some blood congestion in between tubules and small area of hemorrhage in the interstitium were noticed.

J. Appl. Sci. Res., 8(11): 5459-5469, 2012

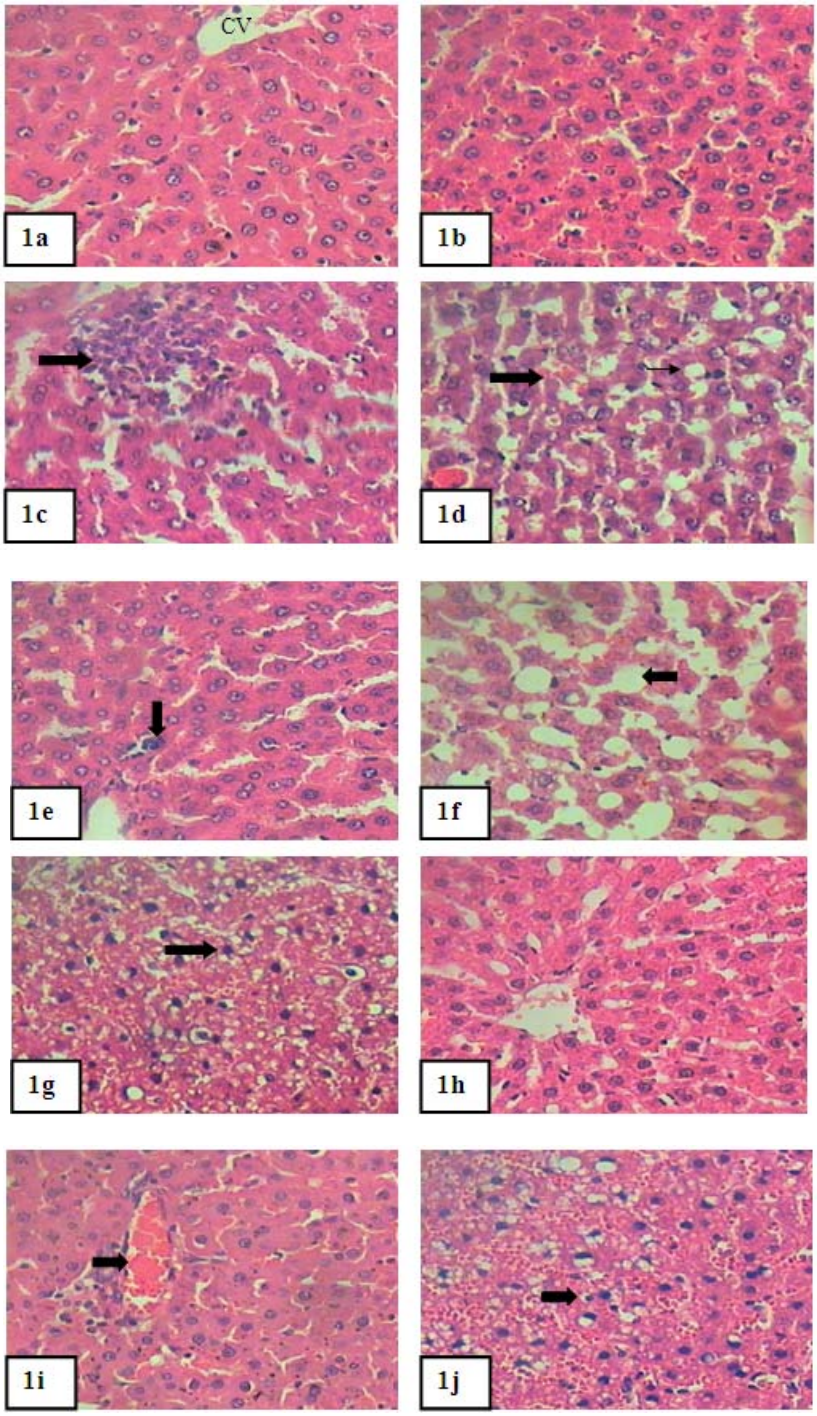

Fig. 1: Histopathological examination of liver sections of male albino rats as affected by oral administration of tested systemic pesticide for 30 days.

Fig. (1a): In the control untreated rats group; Fig. (1b): In rats group treated with low dose (equivalent to 1/20 $LD_{50\%}$) from dimethoate; Fig. (1c) and Fig. (1d): In rats group treated with high dose (equivalent to 1/10 $LD_{50\%}$) from dimethoate; Fig. (1e): In rats group given an oral low dose (equivalent to 1/20 $LD_{50\%}$) of carbofuran; Fig. (1f) and Fig. (1g): In rats group given an oral high dose (equivalent to 1/10 $LD_{50\%}$) of carbofuran; Fig. (1h): In liver sections of rats group received orally the low dose (equivalent to 1/20 $LD_{50\%}$) of carbendazim; Fig. (1i) and Fig. (1j): In liver sections of rats group received orally the high dose (equivalent to 1/10 $LD_{50\%}$) of carbendazim.

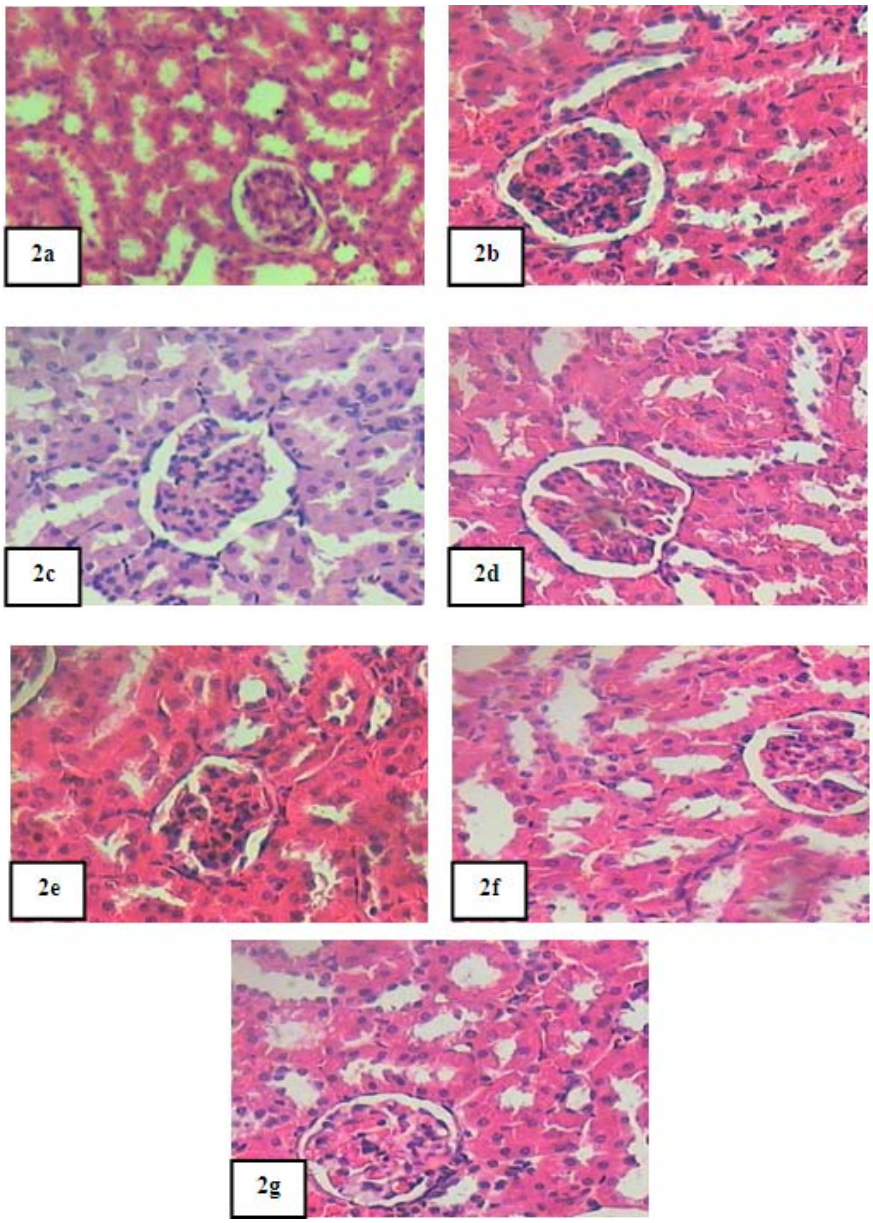

Fig. 2: Histopathological examination photomicrographs for kidney sections of male albino rats as affected by the oral intake of tested systemic pesticides for 30 Days.

Fig. (2a): Kidney section of the control untreated rats group; Fig. (2b): kidney section of rats group treated with low dose (equivalent to 1/20 $LD_{50\%}$) from dimethoate ; Fig (2c): kidney section of rats group treated with high dose (equivalent to 1/10 $LD_{50\%}$) from dimethoate; Fig. (2d): kidney section of rats group treated with low dose (equivalent to 1/20 $LD_{50\%}$) from carbofuran; Fig. (2e): kidney section of rats group treated with high dose (equivalent 1/10 $LD_{50\%}$) from carbofuran; Fig. (2f): kidney section of rats group treated with low dose (equivalent to 1/20 $LD_{50\%}$) from carbendazim; Fig.(2g): kidney section of rats group treated with high dose (equivalent 1/10 $LD_{50\%}$) from carbendazim.

Discussion

The extensive use of different pesticides in agriculture and for public health purposes has led to drastic effects in many non-target species including man (WHO/PCS, 1996; Chantelli-Forti *et al*., 1993 and Chaudhuri *et al*., 1999). The current study was performed to investigate the biochemical and histopathological effects of the

commonly used systemic pesticides including dimethoate, carbofuran and carbendazim on liver and kidney of adult male albino rats.

The results of the current study revealed that there were significant decrease in body weights of rats treated with tested systemic pesticides; especially with high dose treatment. These results are in a good agreement with those found by many authors. In this concern, Pant *et al*. (1995) observed a significant decrease are in body weight of rats treated with 0.2 – 0.8 mg carbofuran kg^{-1} body weight. Sharma *et al*. (2005) found that a significant decrease in the body weight gain at high dose (90 mg/kg/day) of chloropyrifos. In contrast, Brkic *et al*. (2008) found that a statistically significant increase in body weight gain of male rats treated with carbofuran at dose 400 mg/kg.

Also, the present data showed that a significant increase in the relative liver and kidney weights of treated rats with high dose of pesticides. The animals treated with low doses of these pesticides showed non-significant increase in the relative weights of liver and kidney compared with control animals.

In toxicological studies, organ and relative organ weights are important criteria for evaluation of organ toxicity (Timbrell, 2000 and Crissman *et al*., 2004). The explanation of liver and kidney enlargement, and their weight increment in rats treated with pesticides could be due to the accumulation of abnormal cells. Triglyceride accumulation was the result of an imbalance between the rate of synthesis and the rate of release of triglyceride by the parenchymal cells into the systemic circulation (Plaa, 1975). These results agree with those obtained by Hazarika and Sarkar (2001) who reported that the anilofos induced increase in liver and kidney weight of male rats orally administered 50, 100 or 200 mg/kg of anilofos daily for 28 days. Also, Baklan and Akta (2005) found that significant increase in relative liver weight of male and female rats administered at 200 mg benomyl / kg body weight for 15 days. In contrast, Selmanoglu *et al*. (2001) indicated that rats treated with 600 mg/kg per day carbendazim induced a significant decrease in the liver/body weight ratios. Also, Farag *et al.* (2006) reported that no significant difference were found in the relative liver and kidney weights of rats treated with 28 mg dimethoate / kg body weight / day.

The liver functional transaminases (AST and ALT) and alkaline phosphatase (ALP) enzymes activity in serum are most frequently measured for diagnosis of liver diseases particularly infective hepatitis, alcoholic citrrhosis, biliary obstruction, toxic hepatitis and liver cancer (Varshneya *et al*., 1988; Kaneko, 1997; Davidson and Sittman, 1999; Zaahkouk *et al*., 2000 and Abdel-Wahab *et al*., 2007). It has been reported that the normal levels of the former liver functional enzymes activity are up to 37 IU/L for the AST, up to 41 IU/L for the ALT and 68-400 IU/L for ALP in the healthy individual rats serum (Murray *et al*., 1991 and Kaneko, 1997); these levels may be exceptionally elevated into 5-20 times of the previous normal levels in the case of the injured organs. The former liver functional enzymes are not secreted into the blood, any elevation of their activities in blood is resulted from leakage of liver damage cells and from the disturbance and dysfunctions in lever functional enzymes (Murray *et al*., 1991; Kaneko, 1997; Abu-Zeid, 2001 and Attia and Nasr, 2009).

Biochemical analysis results in this research showed that the oral intake of tested systemic pesticides cause an exceptional rise in liver functional enzymes (AST, ALT and ALP) activities in serum of treated experimental animals. The elevation rate in the formed tested liver functional enzymes was increased significantly with increasing the oral intake dose and with extending the oral intake period of tested pesticides.

These results are in approximately similar with the previous study results of Abu Zeid (2001) who reported that the activity levels of GOT and GPT were significantly increased by carbofuran pesticide for 16 days. Selmanoglu *et al*. (2001) indicated that 300 and 600 mg/kg per day carbendazim affected the liver parameters. Soufy *et al*. (2007) found that significant increase in ALP levels after the 3^{th} when monosex tilapia fish exposure to 1/10 $LC_{50\%}$ of carbofuran for 8 weeks. Attia and Nasr (2009) reported that treatment with dimethoate at 75 mg/kg (1/4 LD_{50}) for 28 days caused significant increase in activity of AST and ALT.

The current results showed also that a significant increases in serum of experimental animals treated with tested pesticides; especially in those treated orally with the high dose of pesticides, when compared with the control group. Uric acid and creatinine are useful in early deduction of nephrotoxicity induced by exogenous compounds. These parameters are used as index of renal damage in living organisms (Coles, 1986). Elevation of urea and creatinine concentration in serum of treated male albino rats may be attributed to reduction in glomerular filtration in the kidney and also reflect dysfunction of the kidney tubules (Hayes, 1989 and Walmsley and White, 1994).

These results are in coincidence with those previously obtained by (El-Said *et al*., 1999 and Radwan *et al*., 2001) they found that urea and creatinine concentration were increased in experimental animals after exposure to organophosphorus and carbamate pesticides.

The histopathological examination results in this study demonstrated that 30-day the oral intake exposure of rats to dimethoate, carbofuran and carbendazim at the tested high dose equivalent to 1/10 LD_{50} resulted in degenerative changes in the liver including a large vacuoles in the hepatocytes, hepatic sinusoids congestion; disturbance of hepatic lobules, congested blood vessels in portal area, inflammatory infiltration microvesicle and pyknotic nuclei.

These results are in agreement with many authors; Selmanoglu and Akay (2000) they reported similar histopathological changes including mononuclear cell infiltration, congestion, hydropic degeneration and hepatocellular damage in the liver of male rats treated with dimethoate, endosulfan and carbaryl. Also, Sharma *et al*. (2005) who found that a 30-day exposure of male rats to technical grade dimethoate at doses of 6 and 30 mg/kg caused portal inflammation, centrizonal congestion and focal hepatocyte necrosis in the liver of rats. Also,

The histopathological examination photomicrographs of the kidney tissues in rats treated with the tested low dose of either; dimethoate, carbofuran or carbendazim showed a normal structure of the renal corpuscles and tubules. While, the photomicrographs of kidney sections in rats treated with the oral high dose (equivalent 1/10 $LD_{50\%}$) of either tested pesticides showed that there were some histopathological alterations in kidneys of treated rats including some blood congestion in between tubules and small area of hemorrhage in the interstitium.

These results are in agree with Khogali *et al*. (2005) found that main changes such as blood congestion in between the tubules. in the kidney of mice treated with 60 mg/kg dimethoate pesticide.

In general, it could be concluded that liver in mammals and experimental animals is more effective and injured from the oral intake of tested systemic pesticide than kidneys may be the solubility of these pesticides in water and therefore they easily excreted in urine and no accumulation in kidneys resulting in less injury for than liver which more affected by these pesticides.

In conclusion, the results of the current study indicate that dimethoate, carbofuran and carbendazim induces dose-dependent biochemical and histopathological changes in the liver and kidney of exposed rats. According to these results, it is suggested that systemic pesticides exposure might cause hazardous effects ; especially at high doses to non-target organisms, including humans. Careful attention should therefore be given for a long period during its field application to avoid possible adverse effects to consumers, who are increasingly aware of the potential for contamination of food and drinking water.

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