

STUDY THE BEHAVIORAL CHANGES AND GRAVIMETRIC CHANGES FOR WEIGHT ORGANS IN LIVER, KIDNEY AND SPLEEN EXPOSURE TO INSECTICIDE IMIDACLOPRID IN THE WHITE MICE.

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ABSTRACT

This study was conducted to determine the effect of different concentrations of the insecticide Imidacloprid (IMD) laboratory for the first time in Iraq on male Swiss mice aged 8-12 weeks with a body weight ranged between 20-25 g, and noted the different effects of concentration on the behavior of the mice and the weight changes for organs liver, kidney and spleen, the effects have been examined after 14, 28 days after administration of (0.06, 0.12 and 0.18) mg/kg from body weight. from the results it has been observed that there was a reduction in the animals' movement and activity and clear reduce for pellet and water intake; Also we observed significant decrease ($p \leq 0.05$) in the weight of organs liver, kidney and spleen, results recorded less weight for liver (0.720 ± 0.04), kidney (0.140 ± 0.024) and

spleen (0.40 ± 0.005) after 14 days of oral administration with insecticide (IMD) at dose 0.18 mg/kg.bw.

KEYWORDS: Imidacloprid, white mice, behavioral, gravimetric changes.

INTRODUCTION

Pesticides are used extensively throughout the world. There are several definitions of pesticide, the Food and Agriculture Organization of the United Nations (FAO) defines pesticide as any substance or mixture of substances intended for preventing, destroying or controlling any pest, including vectors of human or animal disease, unwanted species of plants and animals causing harm during or otherwise interfering with the production, processing, storage or marketing of food, agricultural commodities, wood and wood products,

animal food stuffs or which may be administered to animals for the control of insects, arachnids or other pests in or on their bodies.^[1]

Exposure to pesticides is one of the most important occupational risks among farmers in developing countries.^[2] Occupational exposure to pesticides is of great interest in order to identify the hazards of pesticide use and the establishment of safe methods of pesticide handling.^[3] This is because pesticide misuse in various sectors of the agriculture often has been associated with health problems and environmental contamination worldwide.^[4, 5]

It is more evidence that many pesticides produce their acute toxic action by activation or inhibition enzymes.^[6] In addition, chemicals transmitted through food chain have harmed to physiological mechanisms. The extensive use of insecticides has been criticized in recent years due to their persistence in the environment and their accumulation in the living tissues of organisms.^[7]

Imidacloprid (IMD) 1-(6-chloro-3-pyridylmethyl)-N-nitroimidazolidin-2-ylideneamine) belongs to a relatively newer group of insecticides, the neonicotinoids. The neonicotinoids are major class of insecticides developed in the past three decades. Neonicotinoids are primarily used as plant systemic insecticides. IMD was introduced in 1991 as the first chloronicotinyl insecticide and has been registered in 120 countries for use on termites and households pests.^[8] Because of its selectivity for insects.^[9] IMD is thought to be safer than other pesticides, and it now has the highest production of all insecticides worldwide.^[10] In agriculture, it is most commonly used on rice, maize, potatoes, vegetables, sugar beets, fruits, cotton and hops for control of sucking insects, is especially systemic when used as a seed or soil treatment. IMD is known to act as a nicotinic acetylcholine receptor (nAChR) agonist.^[9]

MATERIALS AND METHODS

1. Laboratory animals

All experiments were performed on 144 young Swiss male mice, their ages ranged between 8-12 weeks with a body weight ranged between 20-25 g. Mice were obtained from the colony of the National center for drug control and researches, the animals were housed in plastic cages (3mice\cage) with a wire grid covers, supported on ventilated racks in the animals house of the department of biology at the college of Science AL-Mustansiriyah University. Mice were fed with standard balanced pellet that contains special dietary supplement to keep normal activity and growth.

2. Administration of insecticide to animals

Used insecticide was imidacloprid (IMD) in the form of commercial formulation (Roundup) manufactured by Ensystem II, Inc, Fayetteville, NC, that obtained from Baghdad markets (specialized with insecticide control). Gave orally administration, distal water (D.W) used to dissolve the pesticide and preparation of different doses when studying the toxicity of pesticide.

3. Experimental design

In these study mice randomly distribution in to fore groups as follows:

➤ First group

This group include 36 mice were given only water and is considered the control animals.

➤ Second group

This group include 36 mice were subdivided in to two groups, the first administered pesticide dose of (0.06 mg/kg.bw) per day for a period of 14 days, and the second administered the same dose per day for a period 28 days.

➤ Third Group

This group include 36 mice were subdivided in to two groups, the first administered pesticide dose of (0.12 mg/kg.bw) per day for a period of 14 days, and the second administered the same dose per day for a period 28 days.

➤ Fourth Group

This group include 36 mice were subdivided in to two groups, the first administered pesticide dose of (0.18 mg/kg.bw) per day for a period of 14 days, and the second administered the same dose per day for a period 28 days.

4. Acceptable daily intake (ADI) of imidacloprid

To reach the acceptable daily intake (ADI) of food is by pesticide imidacloprid (0.06 mg/kg) of body weight of the organism and adopted on this dose as basis for determining the other doses in addition to use as a compared dose.

5. Studying behavioral changes

Subjected all animals of all experiment to watching over the period of the study, recorded abnormal observations for the animals inside the cage as moving, the amount of food and drinking water, compared with control animals.

6. Studying gravimetric changes

The weight of some organs of the following (liver, kidney, spleen) of all animals groups (experimental and control) has been measured by using sensitive balance.

7. Statistical analysis

The Statistical Analysis System.^[11] Was used to effect of different factors in study parameters. Least significant difference –LSD test was used to significant compare between means.

RESULTS

1. Effect of Imidacloprid on behavior and activity of mice.

Our study showed a different doses of administrated IMD in experimental mice have always accompanied with unusual changes in behavior and activity, the primary symptoms is sluggish, less movement and activity, showed some body hair loss, reduction in the body weight, clear reduce for pellet and water intake in comparison with control group, these sings were more obvious in high dose (0.018 mg/kg.bw) in both experimental animals.

2. Gravimetric changes for weight organs

A. Liver weight

The statistical results recorded a significant decrease ($p \leq 0.05$) in the weight of liver at 14 days after administration 1.18 ± 0.09 , 1.15 ± 0.11 and 0.720 ± 0.04 g respectively at doses (0.06, 0.12 and 0.18mg/kg.bw) from IMD in comparison with control group (1.79 ± 0.03 g) . The weights 1.18 ± 0.09 , 1.15 ± 0.11 g at doses (0.06 and 0.12 mg/kg.bw) was showed non-significant difference ($p \geq 0.05$). The weights of liver at 28 days after administration 1.17 ± 0.54 , 1.00 ± 0.03 and 1.12 ± 0.11 g was showed a significant decrease ($p \leq 0.05$) at doses (0.06, 0.12 and 0.18mg/kg.bw) in comparison with control group (1.85 ± 0.05 g), were the weight of liver was showed a significant difference ($p \leq 0.05$) at dose (0.18mg/kg.bw) between 14 and 28 days after administration ;while the decrease in the weights at doses (0.06 and 0.12 mg/kg.bw) was non-significant ($p \geq 0.05$) in comparison between 14 and 28 days as listed in table (1), figure (1).

Table (1): Effect of concentration and days of Imidacloprid (0.06, 0.12, and 0.18) mg/kg.bw in weight of liver

Concentration	Days		LSD value
	14	28	
0.06	1.18 ± 0.09 b	1.17 ± 0.054 b	0.232 NS
0.12	1.15 ± 0.11 b	1.00 ± 0.03 b	0.243 NS
0.18	0.720 ± 0.04 a	1.12 ± 0.11 b	0.261 *
Control	1.80 ± 0.04 a	1.85 ± 0.05 a	0.174 NS
LSD value	0.349 *	0.202 *	---

* (P≤0.05).

- (a, b, c) Means with different superscripts within each row are significantly different (P<0.05).

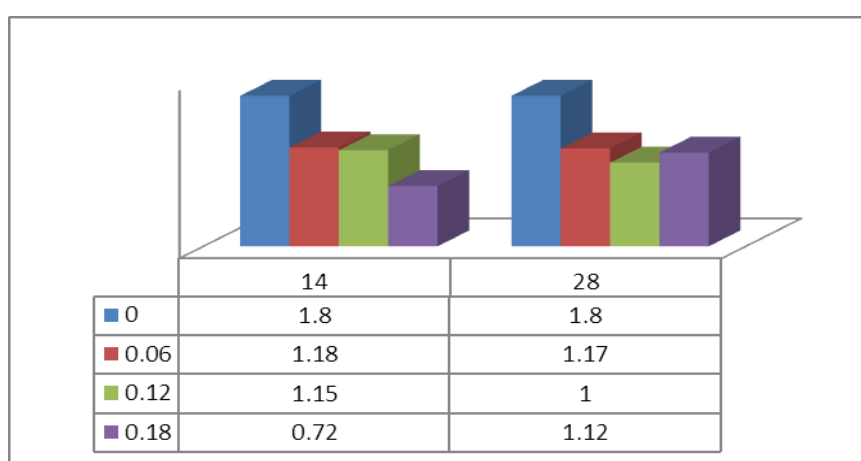


Figure (1): Effects of Imidacloprid doses on liver weights at 14 and 28 days respectively.

B. Kidney weight

The statistical results recorded a significant decrease ($p \leq 0.05$) in the weight of kidney at three doses (0.06, 0.12 and 0.18 mg/kg.bw) after 14 days from administration 0.233 ± 0.015 , 0.287 ± 0.029 and 0.140 ± 0.024 g respectively in comparison with control group (0.60 ± 0.003 g); while the weights at doses (0.06 and 0.12mg/kg.bw) was non-significant ($p \geq 0.05$). At 28 days after administration the weight of kidney decreased in all three doses (0.06, 0.12 and 0.18mg/kg.bw) significant decrease ($p \leq 0.05$) 0.280 ± 0.014 , 0.200 ± 0.018 and 0.240 ± 0.024 g respectively in comparison with control group (0.62 ± 0.004 g); there was a significant difference ($p \leq 0.05$) when comparison between 14 and 28 after administration at doses (0.12 and 0.18 mg/kg.bw) as listed in table (2), figure (2).

Table (2): Effect of concentration and days of Imidacloprid (0.06, 0.12, and 0.18) mg/kg.bw in weight of kidney.

Concentration	Days		LSD value
	14	28	
0.06	0.233 ± 0.015 b	0.280 ± 0.014 b	0.044 NS
0.12	0.287 ± 0.029 b	0.200 ± 0.018 b	0.075 *
0.18	0.140 ± 0.024 c	0.240 ± 0.024 b	0.079 *
Control	0.60 ± 0.003 a	0.62 ± 0.004 a	0.039 NS
LSD value	0.071 *	0.058 *	---

* ($P \leq 0.05$).

- (a, b, c) Means with different superscripts within each row are significantly different ($P < 0.05$).

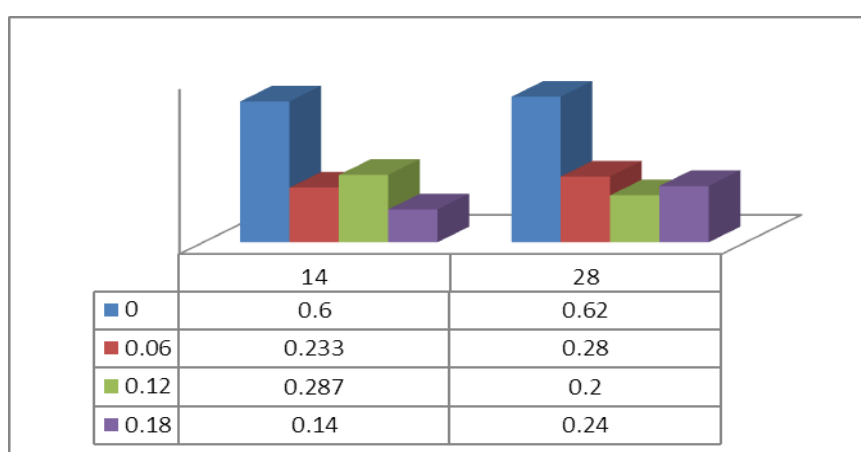


Figure (2): Effects of Imidacloprid doses on kidney weights at 14 and 28 days respectively.

C. Spleen weight

The statistical results recorded a significant decrease ($p \leq 0.05$) in the weight of spleen at 14 days after administration 0.094 ± 0.009 , 0.125 ± 0.025 and 0.040 ± 0.005 g at the three doses (0.06, 0.12 and 0.18mg/kg.bw) in comparison with control group (0.300 ± 0.004 g). The weights at doses (0.06 and 0.12mg/kg.bw) were non-significant ($p \geq 0.05$) at 14 days after administration, the weight at 28 days after administration 0.107 ± 0.006 , 0.137 ± 0.018 and 0.100 ± 0.001 g were showed a significant decrease ($p \leq 0.05$) at the three doses (0.06, 0.12 and 0.18mg/kg.bw) in comparison with control group (0.321 ± 0.15 g); while there was a significant difference ($p \leq 0.05$) between weights at 14 days 0.04 ± 0.005 g and 28 days after administration 0.100 ± 0.001 g at dose (0.18mg/kg.bw) as listed in table (3), figure (3).

Table (3): Effect of concentration and days of Imidacloprid (0.06, 0.12, and 0.18) mg/kg.bw in weight of spleen.

Concentration	Days		LSD value
	14	28	
0.06	0.094 ± 0.009 b	0.107 ± 0.006 b	0.024 NS
0.12	0.125 ± 0.025 b	0.137 ± 0.018 b	0.066 NS
0.18	0.040 ± 0.005 c	0.100 ± 0.00 b	0.013 *
Control	0.300 ± 0.004 a	0.300 ± 0.004 a	0.027 NS
LSD value	0.049 *	0.035 *	---

* (P≤0.05).

- (a, b, c) Means with different superscripts within each row are significantly different (P<0.05).

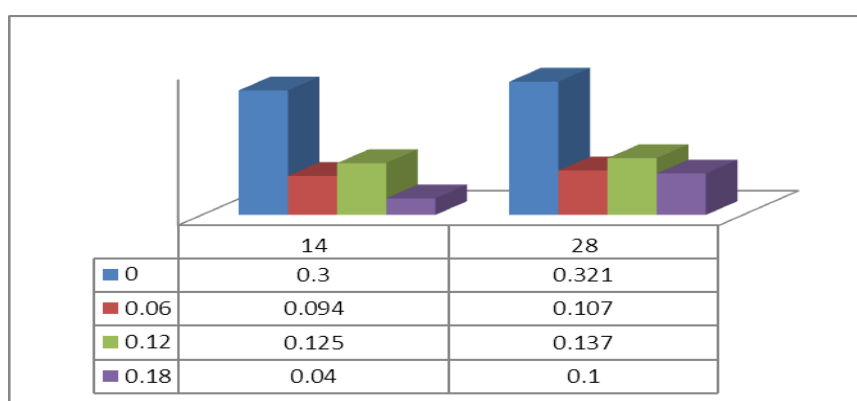


Figure (3): Effects of Imidacloprid doses on spleen weights at 14 and 28 days respectively.

DISCUSSION

This decreasing in the liver weight may be attributed to presence a large number of hepatocytes cells suffer from apoptosis in addition to obtain hepatic necrosis and this results are accordant with the pathological changes in the liver histology and agreement with study of,^[12] that deals with effect of cyfuthrin pesticide in mice for 28 days after administration as the cause of low liver weight after the increase in weight when acute exposure to pesticide for a short period.

From the results that decline in kidney weight for all groups doses in the two periods 14 and 28 days after administration may be attributed to get damage in cells lining of the renal tubules which are accordant with pathological changes in the kidney histology and this results are agreement with study of,^[13] who reported mild congestion in rat kidneys when administration with IMD at 40 mg/kg, also agreed with study of,^[14,15,16] who reported tubular necrosis and degeneration of epithelial cells in renal tubules at rats exposed to carbendazim and cadmium and ethanol respectively, also agreed the decline of kidney weight with study

of,^[17] who showed swelling of the tubules and lining of the bowman's capsules which are probably related to disturbance of the ionic milieu of the cells treated with dimethoate 40 EC.

This decline in spleen weight expose to IMD for all groups doses in the two periods 14 and 28 days after administration may be attributed to damage or death some of spleen cells and reflected on the weight organ and this effects in accordant with pathological changes in the spleen histology, and this study results are agreement with study of,^[18] who observed that the spleen of mice exposed to IMD dose are indicative tissue destruction and injury reflecting IMD induced death of lymphocytes and also showed disintegration of white pulp in the spleen of IMD treated rats with severity lesion at dose 160 mg/kg.bw, these results are accordant with histopathological lesions observed in spleen of rats exposed to 0.21mg/kg.bw of IMD,^[19] also agreed with study of^[20] in decreasing spleen weight when chronic exposure to 1600, 3200 ppm of Acetamidrid insecticide in rats absorbed through food.

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