Effect Of Evisect On Organo-Somatic Index And Pathohistological Changes Of Some Vital Organs In White Mice.

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ABSTRACT

Background: Since the studies and researches regarding Evisect toxicity in humans and animals are sparse, therefore we have a little information on the systemic toxicity of (Evisect) poisoning after oral administration on vital organs alterations and weight

Aim: The aim of this study was to provide the harmful effect of Thiocyclam Hydrogen Oxalate pesticide (Evisect) on the organosomatic indicator and histopathological alternations that may occurred in some vital organs such idney and liver in both female and male mice.

Material & Method: Twenty mice were divided into four groups. Untreated control groups were orally administrated tap water only while the treated groups were received about 100 mg/kg of Thiocyclam Hydrogen Oxalate (THO) orally for a period of 14 days.

Results: Study results showed there were no significant differences (p>0.05) between control and treated groups in organ weights and body weight (organosomatic index) excepting between both treated male and female groups in kidney and liver weights, after 14 days of THO administration. On the other side, the results of histological study for kidney show moderate vascular congestion, mild tubular epithelial degeneration and only mild glomerular shrinkage, no significant inflammation or necrosis seen. The histological study for liver shows diffuse feathery and hydropic degeneration of hepatocytes with lobular disarrangement, congestion and well defined portal chronic nonspecific inflammation with single cell necrosis.

Conclusion: The present research findings reveal that the Evisect insecticide has toxic effects on the kidney and liver histology, although it has no effect on body or organs weight in experimental animals..

INTRODUCTION

The concern has increased in recent years about the excessive and random use of pesticides, due to the environmental pollution that consequent and Harmful health effects on humans. The risks of using these materials have been exacerbated through the sharp rise in their use in agriculture, industry, homeowners and governments¹. Chavarri² and his colleagues were reported that the pesticide residues produced in the product, could cause significant health risks to consumers, who are aware that food and drinking water could be contaminated with these residues, when using pesticides incorrectly. Unfortunately, there has been an excessive use of insecticides among agricultural establishment, without understanding of the potential hazards and possible harmful health effects on the environment, animals and humans. Pesticides are considering as a control agents used to kill undesired insects and other organisms. Until recently, use of these chemicals were not considered as a problem, but On the contrary, it was considered to have a vital function in controlling agricultural farmers³. Evisect the trade name of THO, is a synthetic pesticide contains thiocyclam hydrogen oxalate. It is a nereistoxin analogue insecticide, initially manufactured by Sandoz Laboratories, Basel, Switzerland. Used to kill sucking and chewing insects on a wide range of crops. The chemical name of THO is N, Ndimethyl1,2,3-trithian-5-amine hydrogen oxalate. It was usually prepared as a powder or as soluble granules, and

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its residues do not persist in the environment because it decomposes quickly⁴.

THO (Evisect) at low concentrations acts as acetylcholine receptor stimulator and at high concentrations acts as antagonist without affecting cholinesterase activity. Although it has been well demonstrated in the animal model, it is known to cause liver, kidney, heart, and lung injuries after ingestion⁵. Ware and Whitacre also observed that Evisect act as channel blockers at high concentration. It is a selectively stomach pesticide with contact effect of lepidopterous and coleopterous pests, some dipterous and thysanopterous pests. It has been reported to have many side effects, including skin and eyes irritation, and may results in allergic reactions by skin contact, as well as when it is swallowed it will be harmful and toxic⁶. Evisect used widely in South East region of Saudi Arabia for agricultural purpose. This pesticide is convert into nereistoxin in the insect by the metabolism and interferes with nicotinic acetylcholine receptors. Because the pesticide is an antagonist, it prevents cholinergic transmission, leading to paralysis and insect death⁷. As well as Evisect is use in Saudi Arabia Southwestern region among many pesticides⁶.

MATERIAL AND METHOD.

Thiocyclam hydrogen oxalate (THO):

Thiocyclam hydrogen oxalate insecticide was purchased from local agricultural supplier in Towayreach municipality of Karbala province, which supported as a powder, from (PCI/ India) company. THO was prepared

Vital Organs In White Mice

in concentration (50%w/w, 100gm) then orally administration to experimental animals after dissolved by tap or drinking water. The dosage was 100mg /kg.

Animals

We conducted this experiment at the Applied Medical sciences college laboratory in Karbala University. The male and female mice were housed in separated Cages for at least seven days before the experiment is started, to acclimatize. The housing conditions were an optimum temperature of $24\pm 2C^{\circ}$, the mice were 8-10 weeks old and weighed $21\pm 2g$. The animals were received a pellet diet and water adlibitum.

Organosomatic index

The organosomatic indices of the liver, kidney and testes for the five male mice, and ovaries for five female mice, were calculated to get the organ weight to the body weight ratio of the mice by the following equation: organ weight /body weight x 100.

HS: liver weight/ mouse weight x 100

KS: kidney weight/ mouse weight x 100

- LS: lung weight/ mouse weight x 100
- TS: testes weight/ mouse weight x 100
- OS: ovary weight/ mouse weight x 100

Study design

Twenty experimental animals (mice) were divided into four groups containing five mice each group.

- 1) First group: five male mice used as control group were received tap water only for 14 days.
- 2) Second group: five male mice were received 100 mg/kg THO for 14 days.
- 3) Third group: five female mice used as control group were received tap water only for 14 days.
- 4) Forth group: five female mice were received with100 mg/kg THO for 14 days.

Histological study:

Kidney and liver organs were putted in 10% formal saline to fix it for 24 hours then washed by tap water and embedded in paraffin. By using a rotary microtome, fourmicron thickness sections were prepared and stained with hematoxylin and eosin dyes. The specimens were observed under light microscope; the images were diagnosed for histological study⁸.

Statistical analysis

Statistical comparison was performed by using ANOVA table (a one-way analysis of variance) followed by Student's t-test, using SPSS computer program. All results were expressed as mean \pm SD for five animals in each group using the low significant difference (LSD), P values of <0.05 were considered for comparison of data in control group and treated groups, to indicate statistical significance.

RESULTS

Organosomatic index:

The results of organosomatic indicator in the beginning of experiment in compared with organosomatic indicator in the end of experiment, are presented in Table (1). The organosomatic indicator of both male and female mice of control groups compared with both male and female of treated groups for lung organ, shows there were no significant differences (p>0.05) between them. While there was a significant difference (p<0.05) between treated male and treated female groups only, in organosomatic indicator for liver organ. The results show there was a significant difference (p<0.05) between both treated and control male groups, and treated female group in kidney organosomatic indicator. On the other hand, there was no statistical difference (p>0.05) between testes organosomatic indicator of both treated and untreated male groups. In addition, there was no difference in ovary organosomatic indicator between female control group compare with treated group, which administrated 100 mg/kg of Thiocyclam hydrogen oxalate orally. These results revealed to, that pesticide (THO) has no effect on weight of body or organs in experimental animals.

Table (1): Organosomatic index Mean for lung, liver, kidney, testes and ovary organs of mice, which received 100 mg/kg for 14 days.

Q-S index Group	Lung M±SE LS	Liver M±SE HS	Kidney M±SE KS	Testes M±SE TS	Ovary M±SE OS
Control male	0.90±0.04ª	6.65±0.34ª	0.80±0.04ª	0.35±0.03ª	-
Treated male	0.91 ± 0.07^{a}	7.29±0.42 ^{ab}	0.81 ± 0.05^{a}	0.35±0.02ª	-
Control female	1.07 ± 0.08^{a}	6.52±0.12 ^{abc}	0.68±0.01 ^{ab}	-	1.21 ± 0.08^{a}
Treated female	0.88±0.09ª	5.45 ± 0.94^{ac}	0.65±0.02 ^b	-	0.20±0.05ª
O-S index: organosomatic Index SE: standard error The different small letter refer to significant differences (p<0.05)					

Vital Organs In White Mice

Kidney: the findings of pathohistological study of kidney's tissue shows normal renal tubules, glomeruli and no significant vascular congestion or interstitial inflammation in control group as in Fig (1). Compare with THO treated groups that shows moderate vascular congestion, mild tubular epithelial degeneration and only mild glomerular shrinkage in both male and female mice, which received100mg/kg as in Fig (2) and Fig (3). On the other side, there was no significant inflammation or necrosis seen in the tissue of kidney.

Liver: the pathohistological study results of mice liver tissue shows preserved lobular architecture of hepatic plates with no vascular congestion and portal inflammation as in Fig (4). Compare with liver tissue of treated group, which shows diffuse feathery and hydropic degeneration of hepatocytes with lobular disarrangement, congestion and well-defined portal chronic nonspecific inflammation with single cell necrosis, these changes occur in male and female mice as in Fig (5) and Fig (6).

DISCUSSION

Cartap (thiocarbamate), bensultap, thiocyclam and thiosultap are represent the currently used analogues of Nereistoxin. Evisect is a natural substance; inhibit primarily the ion channels of postsynaptic nicotine acetylcholine. It was extracted from Lumbriconereis heteropoda⁹. The present study result is agreement with EPA about organosomatic indices, when he proved that the pesticides did not affect neither decrease nor increase on the weight of organs, but it may cause alternations in the tissue of these organs led to tumors¹⁰. Also these results were disagreement with the Arfat and his team findings, who reported that the effect of imidacloprid a neonicotinoide pesticide on male mice led to decreased the weight of body accompanied with high toxicity symptoms¹¹. Pesticides have a negative effect on the tissues when using at high doses and can cause tumors, cell membrane damage and the local necrosis of the liver, pancreas and kidney organs¹². The results of this study are agreement with those of Abdelmaged and his colleagues that showed the Evisect caused damage in the liver despite the absence of changes in AST, ALT and ALP activities. The results of such slight to moderate degeneration in liver cells and necrosis with scattered foci infections were observed. This means that the liver is still able to work in spite it was showed the breadth portal cords and a slight hyperplasia in the portal tracts. In a related study the results show shrunk glomeruli, breadth of Bowman's space and the tuft lobular was hidden in kidney's tissue¹³.



Fig (1) shows kidney of control group (400x H&E)



Systematic Reviews in Pharmacy

Vital Organs In White Mice Fig (2) shows kidney of treated male group congestion (black arrow) (100x H&E)



Fig (3) shows kidney of treated female group shrinkage (black arrow), degeneration (red arrow) (100x H&E)



Fig (4) shows liver of control groups (100x H&E).



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Liver treated male group, congestion (black arrow), (Degeneration red arrow) (100X H&E).

Effect Of Evisect On Organo-Somatic Index And Pathohistological Changes Of Some



Fig 6 shows Liver of female treated group (100X H&E).Observation

The phenotypic changes of some organs of the mice that received thiocyclam hydrogen oxalat were observed in end of experiment. The pancreas organ of treated mice was swollen, changed in color to yellow, then to green, and sometimes it was exploded. Also the effect of THO pesticide on liver organ was represented in difficulty to carry it by hand because it was being fractionated into pieces. The kidney was also swollen as an abnormal change.

Conclusion: This study proved there are histopathological alternations in kidney and liver of mice which received THO pesticide. That mean the liver and kidney were the most organs affected by toxicity of THO. Also this study confirmed there is no different between organosomatic index of sex organs and lung in both male and female mice except in treated groups for both sex in liver and kidney. So more studies about the pesticides that used in agriculture, are needed in different animals especially in human to demonstrate its toxic and harmful impacts in different organs.

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