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TERATOGENIC EFFECTS OF ABAMECTIN AND TRALOMETHREN ON ALBINO RAT

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ABSTRACT: *Two pesticides, Tralomethren and Abamectin were used to treat albino rats, Rattus norvegicus to clear their teratogenic effects.*
Lethal doses that kill 50% of rats came to be 13 mg/kg b.w and 23 mg/kg b.w of Tralomethrien and Abamectin respectively.
Doses of 1/10 and 1/30 of LD50 were introduced orally by using stomach tube. Pesticides were introduced starting from 8th to 12th day of gestation.
Both pesticides caused pronounced deleterious effects on all studied parameters
There were increases in resorbed foetuses as it reached ten times as control, upon treatment.
Foetuses' weight and length showed lower values than untreated. Mean weight of foetuses' that produced from Tralomethren treated parents were 2.95 and 3.2 gm for 1/10 and 1/30 LD50 respectively.
Where as it came to be 2.5 and 3gm for Abamectin treated parent with 1/10 and 1/30 LD50 respectively.

Key words: *teratogenic, abamectin, tralomethren, albino rat.*

INTRODUCTION

Both Abamectin and Tralomethrien are on great importance in agriculture. Concentrations of pesticides are increasingly magnified in tissue and other organs along the food chain (Morita *et al* 1975, Mori *et al* 1983 and Abdel-Gawaad and Shams El- din 1990.

Different pesticides have different effects on ecosystem. Many of these effects are often not noticed as causing cancer ,tumors, reproductive failure ,cellular and DNA damage and teratogenic effects ,though we can not generalized those effects . Beside, Ecological damaging effects of pesticides extended beyond individual organism to ecosystem.

Fortunately, there is now concern about environment and in some instances human health effects of excessive use and abuse of pesticide.

The purpose of our study was to investigate to concentrate on the teratogenic activity of both Abamectin and Tralomethrin on the albino rat, as an alarm routine test for these classes of pesticides.

MATERIALS AND METHODS

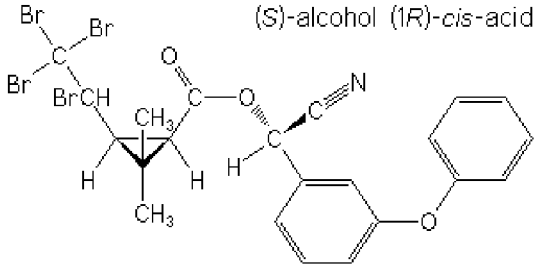
Test animals:-

Laboratory strain of albino rat *Rattus norvegius* was used in this study. They were 2.5 to 3 months old and 140-200 gm weight. The animals were housed in metallic cages under room conditions supplying with enough balanced diet and water and good continuous observation was taken until use.

Pesticides used:

Two pesticides were used:-

A- Tralomethrin:-
Common name, Tralomethrin
Trade name , Tralox
Formulation, 3.5% EC
Chemical name: cyano (3-phenoxyphenyl) methyl 2-2-dimethyl-3-(1, 2, 2 tetrabromoethyl) Cycloproancaboxylate.



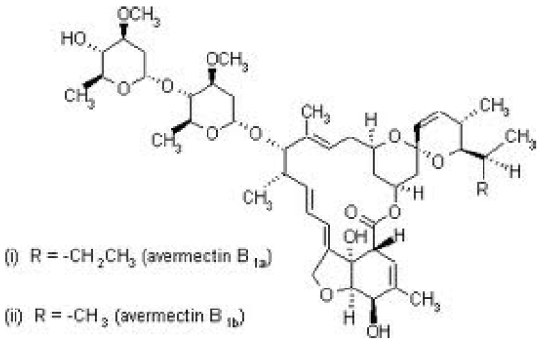
B- Abamectin:-

Common name, Abamectin

Trade name , vertimec

Formulation, 1.8% EC

Chemical name: 5-0-dimethyl-25-di (1-methylpropyl)-22-23 dihydro -25-(1-methyletheyl) avermectin A1a-22, 23-dihydro avermectin b1b.



Toxicity assays:-

1. Determination of LD50 value:

The pesticides were applied orally and the mortality percentages were calculated and LD50 values were determined and 24 hrs and according to Spearman's formula (1975) as follows:-

M= xk +1/2 d -d∑r/n

Where:

M=Log LD50

XK= Log dose causing 100persent mortality.

d= Log interval of dose

r = sum of the number of animals deed of each of the individual dose

n= number of animals on each of a dose.

2.Teratogenic studies on pregnant female rats and foetuses:

Females were examined by means of vaginal smears for the detection of the oestrus status. If the females were in oestrus phase, vaginal smear was taken to ensure the mating and if the sperm was present, this means that the gestations period had begun (zero day).

Regular vaginal smear examination was done daily for about 6 days to follow up the pregnancy. In addition; the body weight was daily recorded for pregnant females to prove the continues pregnancy showing increasing body weight. Fifteen pregnant females were Selected and divided into 5 groups of 3 females for each compound, the first group is left without any pesticide treatment to be used as a control. The 2nd, 3rd, 4th and 5th groups were treated with 1/10 or 1/30 LD50 of either Tralomethrin or Abamectin. The doses of 1.3 and 0.433mg/kg body weight represented 1/10 and 1/30 LD50 values of Tralomethrin while, the same doses for Abamection were 2.3 and 0.766 mg/kg body weight.

Administrations of tested doses were continued for 5 successive days from the 8th to 12 day of gestation period. On the 21st day. The females were scarified and tested for teratoginical examination.

Uterus was dissected out and foetuses were collected from each dam. The uterus was examined morphologically to estimate the resorption sites. One second (1/2) of the foetuses was preserved in Bouin's solution for visceral examination. The remaining foetuses were kept in 96% ethyl alcohol for skeletal examination.

3. External morphology examination of uterus

Resorption sites were counted according to (Cook and Fairweather 1968). The count started from the distal end of the left horn of the uterus to the cervix. The uteri were then opened and examined with magnifying lens for tracing the sites of resorption by their impregnation in 100% ammonium sulphide solution for 20 minutes. Resorption sites that appeared as black spots were easily detected and counted.

4.Visceral examination of foetuses

The foetuses were kept over night in Bouin's solution before examination. Free hand razor sections were made in Bouin's preserved foetuses using the technique described by (Wilson 1965).

Ears and forelimbs of each foetus were removed; meanwhile a cut was made through jaws and just above ears. After examination of the palate, serial sections were made throughout the head. The slices were placed in 70% alcohol in white plastic spot trey. Sectioning was continued from the region of the shoulder joint through the thoracic and abdominal regions to the kidney region.

The urogenital organs were examined in sites. The sections were then examined using dissecting microscope for any induced abnormalities. Visceral malformations and anomalies of each foetus were recorded .

5. Skeletal examination of foetuses:

The eviscerated foetuses, which were previously kept in 95% ethyl alcohol for a period of 7 days, were stained with Alizarin red (S) as described by Staples and Schnell (1964). Muscles of these foetuses were first digested by 2% potassium hydroxide for 6-24 hours according to size of foetuses. Then were immersed in Mallsch's solution alone for 2 days, then rinsed in cold water and cleaned by successive passage in grade concentration (50, 70, 90 and 100%) of glycerol solution.

Before skeletal examination, they were transferred to 70% ethanol, then examined under dissecting microscope.

RESULTS AND DISCUSSION

The results showed clearly the adverse effects of tested pesticides, Tralomethrin and Abamectin on the pregnant female rats and subsequently the other domestic animals. The sites involved in this study were implantation sites, resorption, foetol weight and length. The results are summarized in Table (1) and illustrated in Fig (1). No differences were noticed with respect to the effect on implantation and resorption. No differences were noticed.

Moreover, lengths of produced foetuses were loss than control 4.2 cm.

The recorded values were 2.87, 3.5cm and 2.55, 3.26 cm upon Tralomethrin and Abamectin treatment with 1/10 and 1/30 LD50 respectively.

It appeared that higher concentration of tested pesticides 1/10 LD50 caused cleft palate and hard effects on heart, malformation in skull, ribs stern brae, coccygeal, vertebrae, fore and hind limps.

This study was carried out to explore the expected teratoginc activity of Tralomethrin and Abamectin in rats and hence to other domestic animals wich may be encountered these compounds accidentally .Tralomethrien and Abamectin were administrated orally by a gavages to pregnant female rats using 1/10 and 1/30 of the LD50 of each, respectively.

Pregnant females were treated daily from the beginning of the 8th day until the 12th day of gestation period.

The effect of two compounds on the pregnant female rat namely:

Implantation sits, resorption, foetal weight and foetal length were studied. The results obtained are summarized in Table (1) and Fig (1).

Effect on implantation and resorpion :-

No differences were noticed between the control and Tralomethrin and Abamectin treated groups regarding the number of implantation site (Table 1).

Concerning the resorption foetuses , Abamectin treatment at 1/10 of theLD50 increased obviously the resorption site comparing with the untreated control. The percentages of resorbed foetuses were:

4.33, 29.41, 21.81, 42.18 and 22.35 % for untreated, Tralomethrin an Abamectin at 1/10 and 1/30 of the LD50 of each, respectively.

Table (1): The foetal changes after oral administration of Tralomethrin and Abamectin to pregnant female's rats:-

Treatments	Dose mg\kg B.wt	Mean implantation sites	Resorped foetuses *Mean	%	Foetal weight (gm) *Mean	Foetal length Mean *
Control	-	9.20	0.4±0.35	4.33	3.85±0.24	4.20±0.15
Tralomethrin 1/10 LD50	1.300	8.50	2.5±0.65	29.41	2.95±0.52	2.87±0.42
Tralomethrin 1/30 LD50	0.433	8.25	1.8±0.55	21.81	3.20±0.45	3.50±0.51
Abamectin1/10 LD50	2.300	7.35	3.1±0.60	42.18	2.52±0.36	2.55±0.33
Abamectin1/30 LD50	0.766	8.95	2.0±0.45	22.35	3.00±0.23	3.26±0.52

±standard error

*Measured from the middle of the horizontal line between the two ears till the test coccygeal vertebrae along the vertebral column.

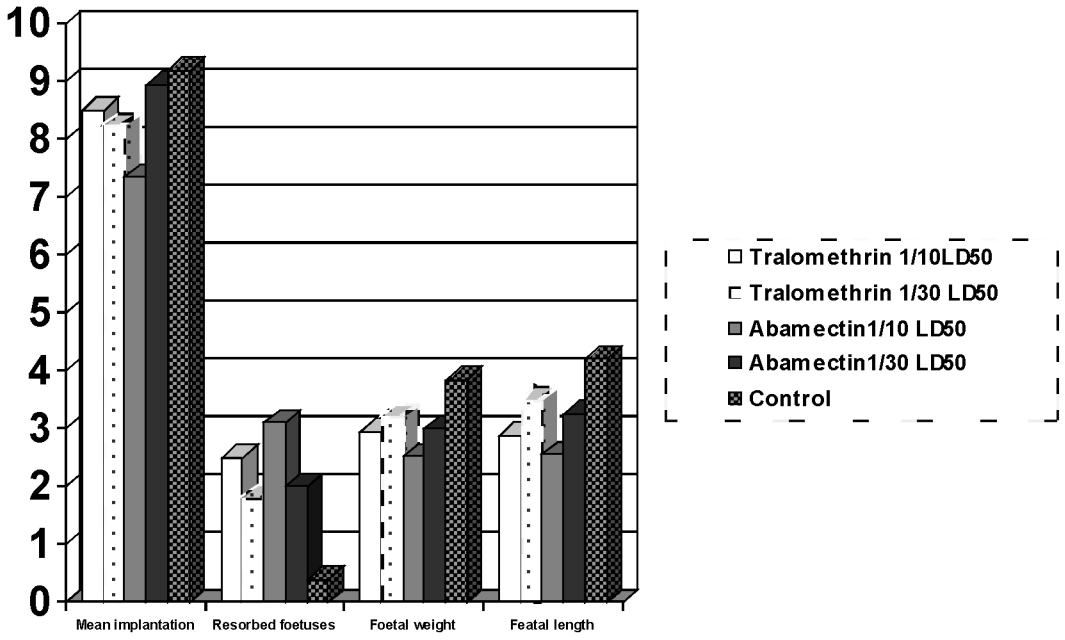


Fig (1): The foetal change of rat foetuses due to Tralomethrein and Abamectin toxicity at 1/10 and 1/30 LD50 of each.

Weight of foetuses descended from the pregnant female rats treated with Tralomethrein and Abamectien at 1/10 and1/30 of LD50 of each were reduced markedly. However, mean weight of foetuses was decreased as dose of compound increase. the mean weight were 2.95 ,3.20 , 2.52 and 3.0 gm for the treatments of 1/10 and1\30 ofLD50 of each Tralomethrin and Abamctin respectively compared with 3.85 gm for that of the control (Table 1) .

Clear differences were found between the foetal length upon Tralomethrin and Abamectin treatments and the untreated control.

The length was decreased from 4.20 cm in the untreated to :2.87 ,3.50 ,2.55 ,and 3.26 cm in case of Tralomethrin and Abamectin treatments at the dose of 1/10 and 1/30 LD50 of each , respectively .

Malformation induced in the general viscera located in head, thorax and abdomen as influenced by the two compounds were recorded and illustrated in table (2) and plates (1,2,3 and 4) . Untreated foetuses showed no malformations either in head and thorax or in abdominal regions.

In head region, the treated foetuses were suffered from cleft plate (palat1). The highest percentage was recorded in the highest dose of Tralomethrin (1/10 of LD50) followed by the highest dose Tralomethrin (1/10 of the LD50). The percentage of foetuses showed cleft palate was: 12.0, 9.1, 25.0, and 8.0 % for the Tralomethrin and Abamectin at 1/10 and 1/30 of the LD50 of each, respectively.

The most prominent effect in thorax region was atrophy of heart (plate 2). The percentage of atrophy cases (table 2) were: 8.0, 4.5, 15.0 and 8.0% for the Tralomethrin and Abamectin at 1/10 and 1/30 of the LD50 of each, respectively.

Tralomethrin and Abamectin treatment at 1/10 and1\30 of LD50 did not exhibit any effects on most organs of abdomen especially on liver and kidney as appeared in plates (1, 2,3 and 4)

Table (2): Visceral malformation in rat foetuses descended from pregnant females treated with different doses of Tralomethrin and Abamectin.

Treatments	Doses mg\Kg B.Wt	No .of examined foetuses	Malformation					
			Head		Thorax		Abdomen	
			Cleft palate		Atrophy of heart		Atrophy of liver and kidney	
			No	%	No	%	No	%
Control	-	35	-	-	-	-	-	-
Tralomethrin1/10 LD50	1.300	25	3	12	2	8	-	-
Tralomethrin1/30 LD50	0.433	22	2	9.1	1	4.5	-	-
Abamectin1 /10 LD50	2.300	20	5	25	3	15	-	-
Abamectin 1/30 LD50	0.766	25	2	8	2	8	-	-

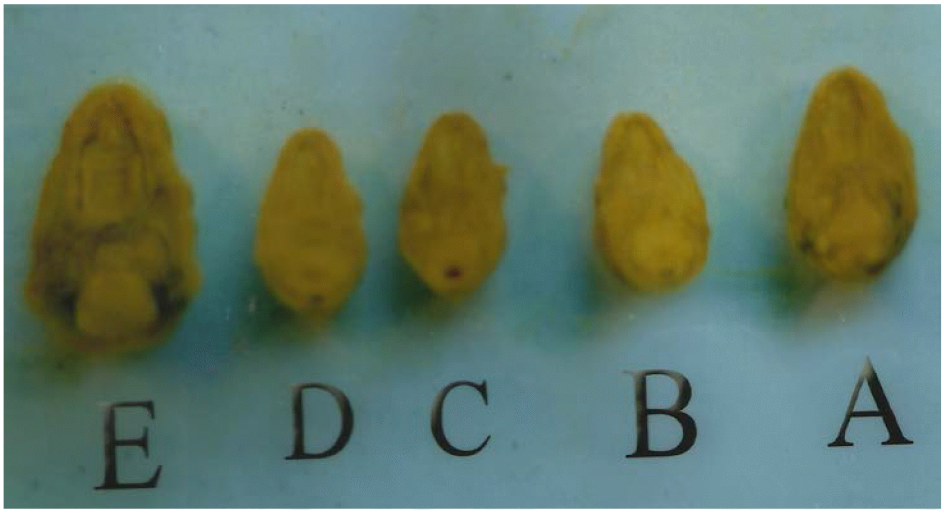


Plate (1): Showing cleft palate:

A. control. **B .Tralomethrin (1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

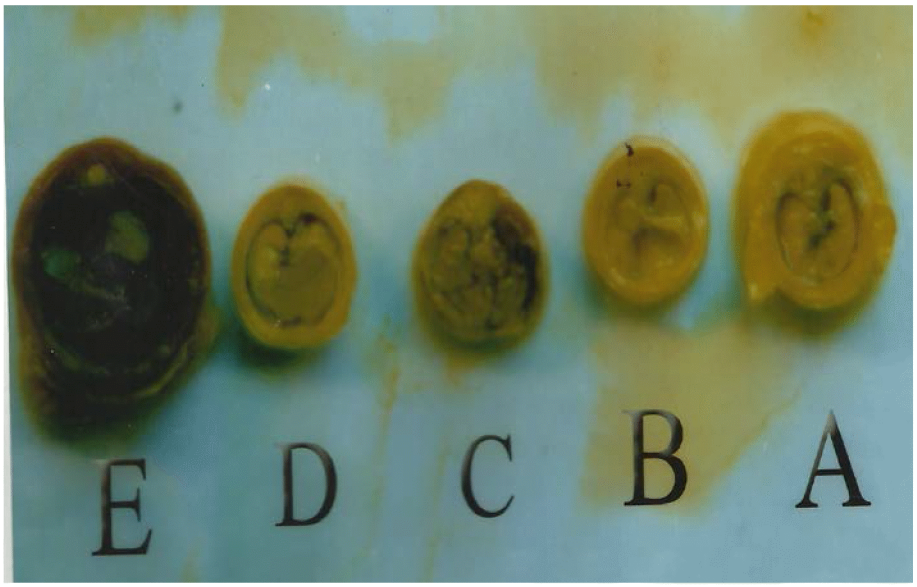


Plate (2): Showing hypoplasia of the heart:

A . Control. **B .Tralomethrin (1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

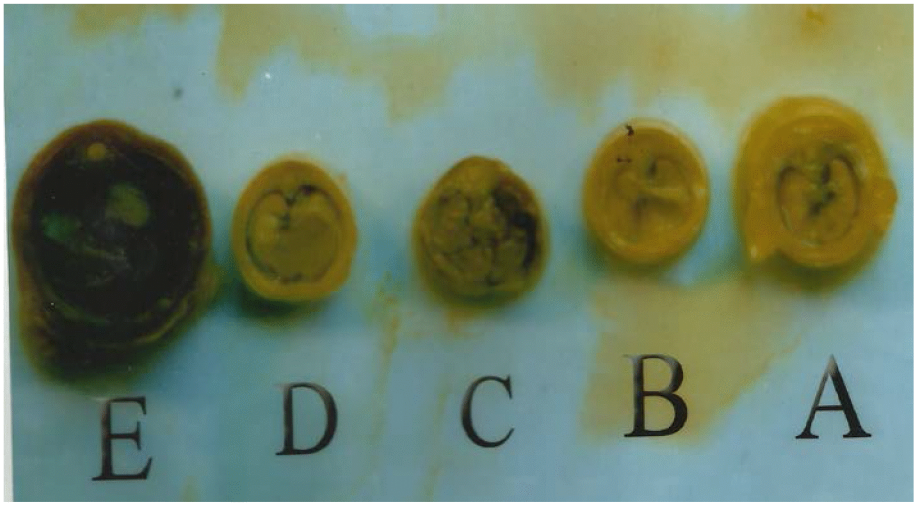


Plate (3): Showing no differences in liver:

A. control. **B .Tralomethrin (1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

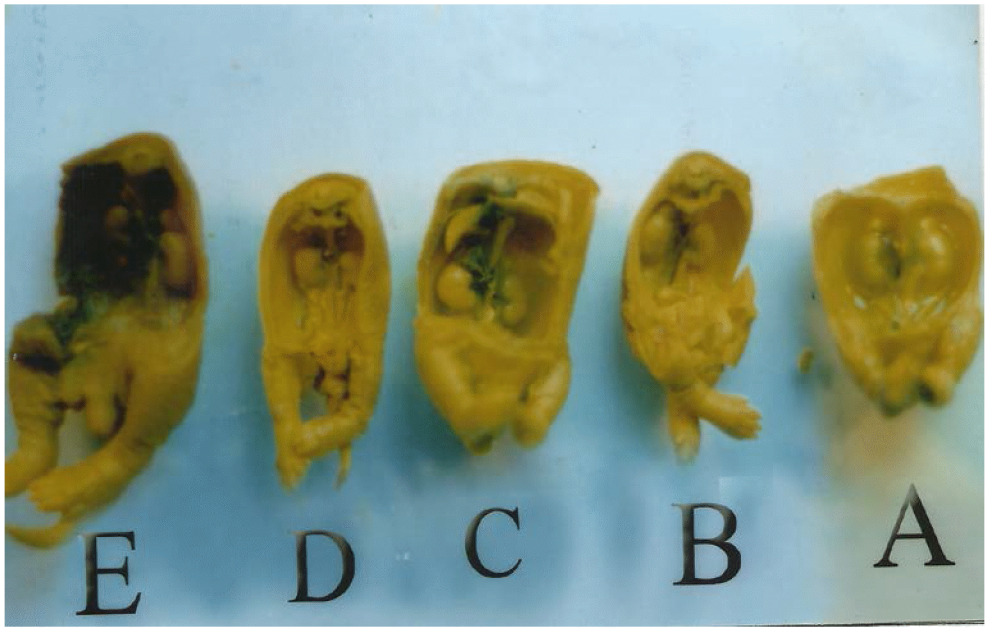


Plate (4): Showing differences in kidneys:

A. control. **B .Tralomethrin (1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W)**
E. Abamectin (0.766 mg \Kg .B.W).

The teratogenic activity of the two compounds tested on the pregnant female rats was more sever on the skeleton than on viscera. The skeleton abnormalities were condensed and more obvious in case of highest dose (1/10LD50) comparing with the other dose (1/30 LD50) . Foetuses of the control were free and have normal skeletal system. Skeletal malformations were in skull, coccygeal vertebrae, sternbrae, ribs, forelimbs and hind limbs. Table (3) and plates (5, 6, 7,8 and 9).

The skull malformation of the foetuses of treated females was absence of cranial bone. The highest dose of Abamectin 1/10 ofLD50 induced 18.18% cranial bone absence as it appeared in Table (3) and plate (5) .The foetuses of the other doses were normal as in the control.

All the tested doses of the two compound caused absence in coccegyal vertebrae. The percentage of absence were 32.0, 11.11, 22.73and15.38 % for the doses of 1/10 and1/3O ofLD50 of both Tralomethrin and Abamectin, respectively. (plate6)

The two tested pesticides showed malformation in sternbrae or in some vertebre of sternbrae. The severity was dose dependent. The percentages of malformed sternbrae were: 12.3, 3.70, 9.09and 3.85%for Tralomethrin and Abamectin 1/10 and 1/30 of LD50 of each, respectively. Plate (7).

Also, the ribs of treated foetuses were obviously affected as shortening in ribs or zigzag ribs. The percentages of wavy ribs were: 20.0, 7.42, 9.09 and 3.85% for Tralomethrin andAbamectin 1/10 and 1/30 of LD50 , respectively. Plate (8).

The severe effect on the skeletal system was the absence of the fore and the hind limbs in some foetuses. The percentage of the individuals showed absence of either fore or hind limbs were: 12.0, 3.70, 9.09 and 3.83 % for Tralomethrin and Abamectin 1/10 and 1\30 of LD50 respectively as appeared in Plates 9 and 10.

Table (3): Skeleton changes in rat faetuses' descended from pregnant female rats treated with different doses of Tralomethrin and Abamectin:

Treatment	Doses mg\Kg B.Wt	No .of examined foetuses	Malformation									
			Skull cranial bone absent		Coccegyl vertebrae absent		Sternbrae absence		Ribs shortage		Fore limbs absence		Hind limbs absence	
			No	%	No	%	No	%	No	%	No	%	No	%
Control	-	30	-	-	-	-	-	-	-	-	-	-	-	-
		25	-	-	8	32.00	3	12.3	5	20.0	3	12.00	3	12.0
Tralomethrin1\30 LD50	0.433	27	-	-	3	11.11	1	3.70	2	7.42	1	3.70	1	3.70
Tralomethrin1\10 LD50	1.300	22	4	18.18	5	22.73	2	9.09	2	9.09	2	9.09	2	9.09
abamectin 1\30 LD50	0.766	26	-	-	4	15.38	1	3.85	1	3.85	1	3.83	1	3.83

Generally, Tralomethrin and Abamectin have a teratogenic activity representing in skeletal and visceral anomalies. Most of skeletal and visceral anomalies found in the present study are common ones. These anomalies are: missing in cleft palate, hypoplasia of the heart, incompletely ossified skull bones, absence of sternbrea, absence of coccygeal vertebrae and absence of limb bones. These malformations are mostly in accordance with those described by several investigations working on other compounds (Hassan *et al.,* 1990; Fouad *et al.* ,1994; Gupta *et al*., 1984 and Sherif *et al.,* 1989). .

Hassan *et al.* (1990), used methomyl for 10 day pregnant rats from the 6^{th} -15^{th} day of gestation .they reported that the used pesticide caused early resorption of foetuses and decreased their weight compared with those of the control. Moreover it caused hypoplasia of heart and liver and caused corrugation of some ribs and absence of sesameoid bones of fore and hind limbs.

Sherif (1991) found that Tamaron treatment increased the mean of the percentage of resorbed foetuses; in Tamaron treatment foetal weight and length were decreased.

El-Ashmaoui and Salah(1994), reached a conclusion that the total number of resorption were significantly increased in treated rats compared with the control , foetal weights were significantly decreased in treated groups reflecting the ability of the tested compound to cause teratogenic effect.

Li *et al.* (1998) reported that examination of foetuses revealed that 3-MC had caused the foetal resorption. The foetuses survived weighted were less than the control foetuses. All rat treated mothers had foetuses with abnormalities, and the main malformations were absence of tail and cleft palate.

Plate (5): Incomplete ossification of skull:
A. control. **B .Tralomethrin(1.30 mg \Kg .B.W) .**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

Plate (6): Absence of some coccygyl vertebrae:
A. control. B .Tralomethrin(1.30 mg \Kg .B.W) .
C. Tralomethrin (0.433 mg \Kg .B.W). D. Abamectin (2.3 mg \Kg .B.W).
E. Abamectin (0.766 mg \Kg .B.W).

Plate (7): Absence of sternbrae:
A. control. B .Tralomethrin(1.30 mg \Kg .B.W) .
C. Tralomethrin (0.433 mg \Kg .B.W). D. Abamectin (2.3 mg \Kg .B.W).
E. Abamectin (0.766 mg \Kg .B.W).

Plate (8): Shortage in ribs:

A. control. **B .Tralomethrin(1.30 mg \Kg .B.W) .**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

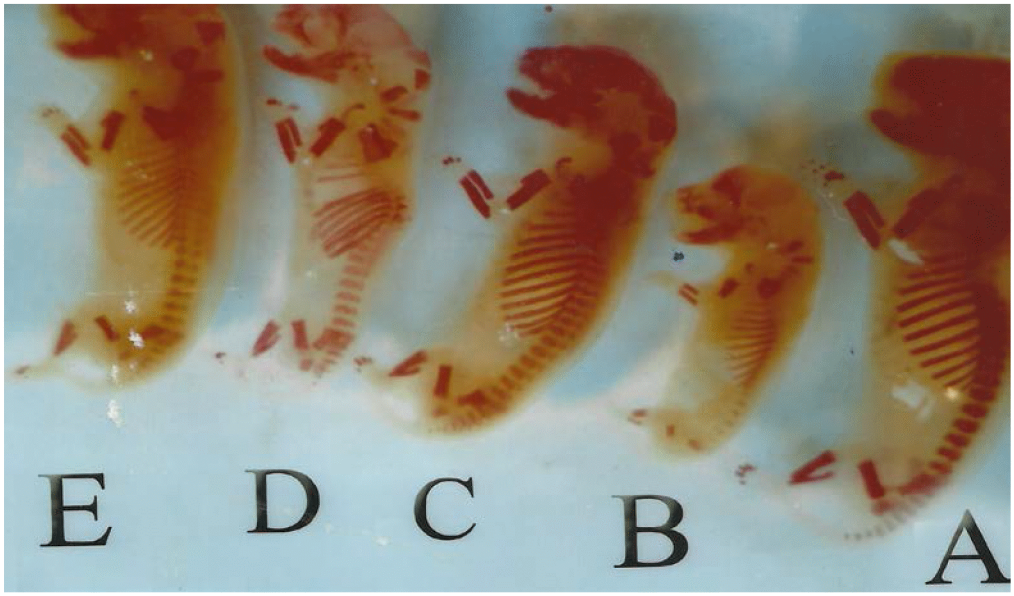


Plate (9): Absence of forelimbs:

A. control. **B.Tralomethrin(1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

Plate (10): Absence of hind limbs:
A. control. **B .Tralomethrin (1.30 mg \Kg .B.W).**
C. Tralomethrin (0.433 mg \Kg .B.W). **D. Abamectin (2.3 mg \Kg .B.W).**
E. Abamectin (0.766 mg \Kg .B.W).

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تأثيرات الأبامكين والترالوميثرين على أجنة الفأر الأبيض

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الملخص العربى

تم استخدام مبيدين حشريين ترالوميثرين وأبامكتين لمعاملة الفأر الأبيض لايضاح تأثيراتهم على الأجنه.

ظهر ان الجرعات المميته ل50% من الفئران كانت 13مجم/كجم و23مجم/كجم لكلا من الترالوميثرين و الأبامكتين على الترتيب

تم استخدام 1/10, 1/30 من الجرعه النصف مميته وتم اعطائها بالفم باستخدام انبوب المعده بدءا من اليوم الثامن لليوم الثانى عشر للشبق .

سببت المبيدات المستخدمه اضرارا واضحه لكل القياسات المدروسه كذالك معدل امصاص الاجنه حتى 10 مرات من الكنترول وكذا قل وزن وطول الاجنه الناتجه من الأباء المعاملة عن الكنترول.