

and 10 cm in height), half filled with tap water using a glass dropper. About 250-300 hatched larvae were reared in each pan. A mixture of bread, dried yeast and dried milk in 2 : 1 : 1 ratio respectively, were ground, sieved, and used as larval food. The amount of food added daily was increased with larval age; excess food was avoided to prevent scum formation. The breeding bowls were aeriated by means of an electric small bubbler. Exuviae with any scum formed were daily removed.

The breeding bowls were covered with muslin cloth as providence against any foreign insects.

Fig.(1)

- The pupae

Pupae were collected daily by means of a wire mesh or a small medicinal dropper and transferred to plastic containers (10X10 cm) half filled with tap water and than introduced inside adult breeding cages.

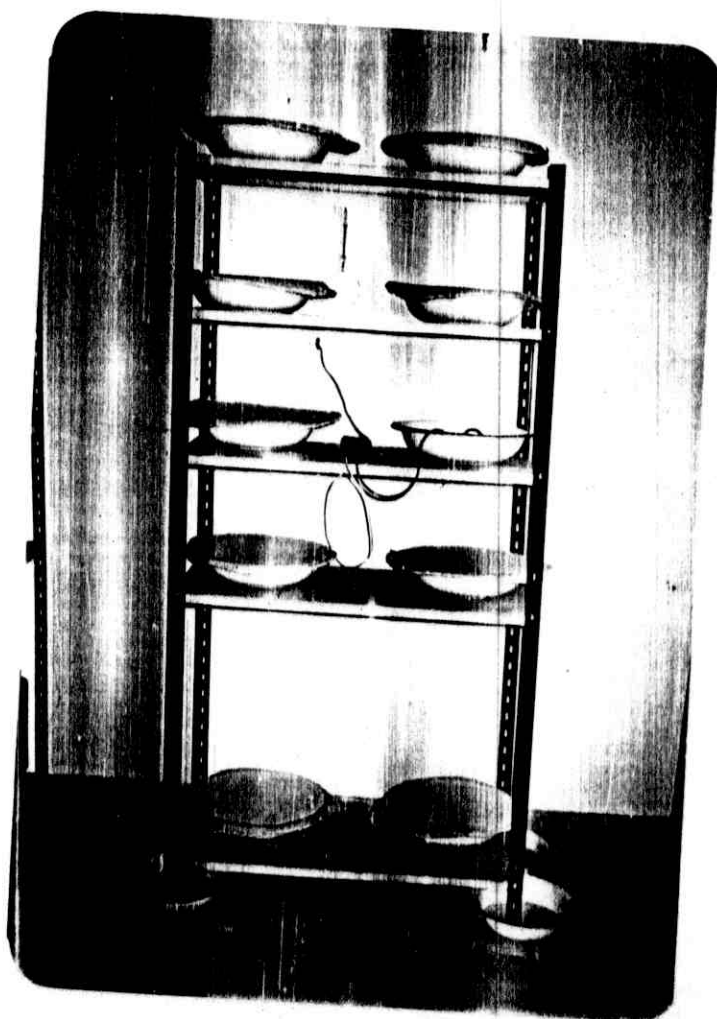
- The adults

Adult mosquitoes were reared in wooden cages (50X50X50 cm.) the floor made of wood, the top and the three sides were covered with wire screen.

Fig.(1)

A side of the insectary shows:

- White enameled pans where larvae of Culex pipiens were reared, covered with muslin and provided with air bubblers.



Females were fed on blood meals through the introduction of a pigeon into the adult cages. During feeding, cages were covered with black cloth to achieve a more rapid feeding. Fig.(2).

Plastic cups half filled with tap water were placed inside the cages for egg oviposition

A sponge soaked with 10% sugar solution was used for feeding males, from time to time the sponge was replaced by a new one to avoid fungal contamination.

(2) Bacterial larvicides

Two mosquito- pathogenic bacteria were used during the present investigation:

- 2.1) Bacillus thuringiensis - serotype H-14
- 2.2) Bacillus sphaericus strain 1593-4

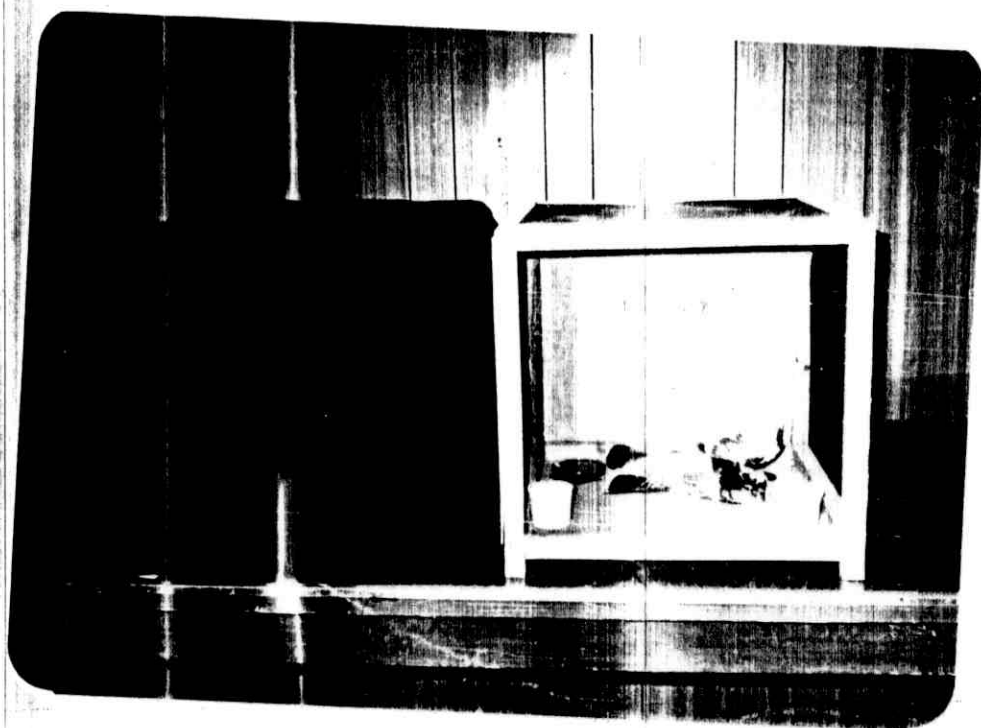
Both bacterial species are belonging to Family: Bacillaceae.

Formulation:

A commercial formulation in the form of a flowable concentrate under the name of "TEKNAR", SAN-402 based on Bacillus thuringiensis H-14, produced by Sandoz Co. and a Bacillus sphaericus strain 1593-4 in the form of

Fig. (2)

An adult rearing cages provided with
peigons for females, feeding (right);
covered with black cloth (left).



a wettable powder produced by SOLVAY- Belgium

(3) Chemical insecticide

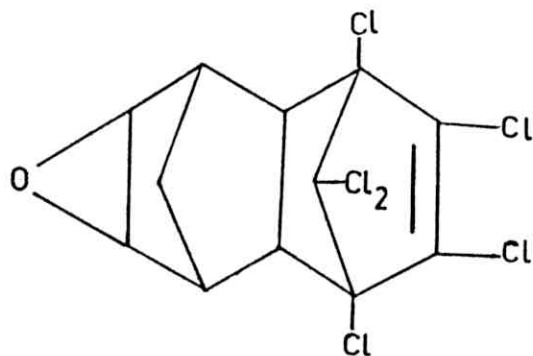
Synonymy: Chlorinated aryl hydrocarbons

(containing 6 or more chlorines)

Common name: Dieldrin

Chemical structure:

Not less than 85% of 1, 2, 3,
4, 10,10-hexachloro-6,7-epoxy- 1, 4, 4a,
5, 6, 7, 8, 8a- octahydro-1,4-endo-exo-
5, 8-dimethanonaphthalene.



Endo-exo $C_{12} H_8 Cl_6 O$

(4) Bioassays

4.1) Preparation of the bacterial concentrations:

Different concentrations of the tested bacterial materials were prepared as follows:

Bacillus thuringiensis serotype H-14.

A suspension was first prepared by adding 1 ml of the commercial formulation flowable concentrate to a volumetric flask containing 1000 ml distilled water. From this stock concentration, serial $1/10$ dilutions were made according to the required concentrations.

In case of Bacillus sphaericus 1593-4, a suspension was prepared by weighing 0.25 mg of dry wettable powder in a volumetric flask containing 50 ml distilled water; from this stock concentrations, serial dilutions were also made in the same manner previously described.

The bacterial suspension was vigorously shaken prior to each use. Dilutions were freshly prepared before each bioassay. Concentrations were expressed in parts per million (PPm) in order to calculate the LC's

4.2) Preparation of the dieldrin concentrations:

Dieldrin concentrations were prepared as follows:-

The insecticide formulation was dissolved in ethyl alcohol and used as a stock solution from which the desired concentrations were prepared.

4.3) Susceptibility experiments:

For testing the larval susceptibility levels to dieldrin and to the bacterial larvicides (Bacillus thuringiensis H-14 and Bacillus sphaericus) , plastic cups (150 ml capacity), each containing 99 ml of distilled water, were used. One ml of the tested larvicide preparation at the desired serial concentration was infiltrated under the water surface with a pipette. A group of 10 larvae of the early 3 rd instar was introduced in each cup . Every dilutions had a 6 replicates.

The test was run at the same temperature and relative humidity as that used for rearing condition. Control experiments were tested similarly following the same procedure previously mentioned. Control tests with more than 5% mortality were discarded.

All cups were once provided with larval food. Dead larvae were not removed from their containers until completion of the experiment i.e. after taking the readings for 24, 48, 72 hours.

Reference cups were made in order to take samples for the histopathological studies.

4.4) Combined effect of bacterial and chemical larvicides:

The joint action of the chemical and each bacterial larvicides, as well as the effect of the two bacterial preparation together were tested. The same technique previously mentioned for the determination of the median lethal levels was followed, instead of using one larvicide, one ml from the concentrations which produced 50 % mortality of each larvicide was added to plastic cup containing 98 ml distilled water simultaneously. Susceptibility experiments of larvae that were pre-treated with the bacteria or the chemical larvicide for 24 hr and then treated with the other larvicides were carried out in six different combinations as follows:-

Pre-treatment

Additional treatment
after 24 hr

1- B.t. H-14

B.sph.

2- B.t. H-14.

DLD

3- B.sph.

B.t. H-14

4- B.sph.

DLD

5- DLD

B.t. H-14

6- DLD

B.sph.

Note:

- (1) B.t. H-14 (Bacillus thuringiensis H-14).
(3) B.sph. (Bacillus sphaericus).
(5) DLD (Dieldrin).

4.5) Mortality readings

Mortality data were recorded 24, 48, and 72 hours after treatment. Percentage of mortalities was corrected by Abbott's formula when the mortality among control experiment exceeds 5%.

4.6) Statistical analysis of the data

Percentage of mortality was converted into probits, which were then plotted against the concentrations. The median lethal concentration (LC_{50}) was determined graphically.

T-test was used to evaluate the effect of all different larvicides either when they were used separately, in mixtures of two preparations or when one preparation was firstly applied then followed by another different preparation 24 hours later.

5) Histopathological studies -

The pathological action of the chemical insecticide (dieldrin) and the bactericides on Culex pipiens larvae was studied.

5.1) Processing of larval samples

Samples were selected from larvae exposed to the bactericides, chemical insecticid or the prepared mixtures, (after bioassay testes), in two different forms:

- 1- Moribund larvae which were hardly responded to the mechanical stimuli.
- 2- Freshly dead larvae (never responded to any stimuli.
- Collection of larvae were made during observation of the treated ones.

5.2) Reagents used.

According to Gurr, E. (1962), the following reagents were used:

1- Bouin's fluid

Recipe:

Picric acid, saturated aqueous	750 ml
Formalin (40%)	250 ml
Glacial acetic acid	50 ml

2- Ehrlich Haematoxyline stain.

Haematoxyline 2% in absolute alcohol	100 ml
Potash alum 2.5% aqueous	100 ml
Glycerine	100 ml
Glacial acetic acid	10 ml

The solutions were mixed well; allowed to ripen in a 500 ml. closed bottle for three months. Filtrated before use.

3- Eosin Stain

Water soluble eosin powder	1 g.
Dist. water	100 ml

5.3) Principle of the technique.

1. Fixation

Selected larval samples were fixed in aqueous Bouin's fixative for 24 hr, washed for 3 successive days in 70% ethyl alcohol to remove the excess of picric acid.

2. Dehydration

The fixed larval samples were dehydrated with serial dilutions of ethyl alcohol starting with 70, 80, 90, 96% and then absolute

alcohol. The samples were passed in each grade for 5 minutes except the last one where specimens passed twice to ensure complete dehydration of the specimens.

3. Clearing

Xylol was used as a de-alcoholising agent. Starting with xylol and absolute ethyl alcohol in 1:1 mixture then passed in pure xylol 2 minutes for each step.

4. Embedding

Paraffin wax with 58 C° melting point was used in the following steps in a hot oven adjusted with the same temp. A mixture of the melted wax and the xylol in (1:1) ratio was firstly used, then the larval specimens were passed twice in pure melted paraffin, 30 minutes each. The larval samples were embedded in wax block and oriented to the desired position. The blocks were allowed to cool, when a heavy film was observed across its top; allowed it to float on cold water in order to prevent crystallisation and get a homogeneous blocks.

5. Sectioning

A rotary microtome was used for sectioning. The block was oriented in the right position against the microtome knife, a continuous ribbon (50 m. thickness) was cut. Serial sections were flattened on glass slides wetted with dist. water, incubated warm on a hot plate at 40 C°, till complete dryness.

6. Staining

The cutted sections were dewaxed by using xylol, hydrated in mixture of xylol +abs. ethyl alcohol, 100, 96, 90, 80, 70, 50, 30% ethyl alcohol, washed in dist. water, 2 minutes for each step. The sections were finally stained using haematoxyline-eosin counter stain technique.

- Firstly the slides were immersed in haematoxyline stain for 5 minutes.
- Differentiated in a mixture of (70% alc.+ drops of 1N HCl) to remove excess haematoxylin from the cytoplasm.
- Washed in tap water.
- Counterstained with eosin for 30 seconds.
- Washed in distilled water, then dehydrated,

cleared, mounted in canda blasam and allowed to dry.

Finally the slides were examined microscopically and photographed.

IV. RESULTS

IV RESULTS

1. Susceptibility Experiments :

For testing the susceptibility of **the 3rd instar larvae of Culex pipiens** to the two bacterial larvicides (Bacillus thuringiensis H-14 and Bacillus sphaericus strain 1593-4), as well as, to the chemical one (dieldrin), a series of bioassay tests were carried out, in which each larvicide was tested individually. Results of this series of experiments are tabulated in (Table 1-3) and graphically represented in (figs. 3-5).

1.1. Bacterial larvicides:

1.1.1. Bacillus thuringiensis serotype H-14 treatments:

The bacterial larvicide (Bacillus thuringiensis H-14) was assayed against the 3rd instar larvae. Mortality resulted after 24 ,48 & 72 hours post-treatments are presented in table (1) as well as graphically illustrated in Fig. (3).

Table (1)

Susceptibility of the early 3rd instar larvae
of Culex pipiens to Bacillus thuringiensis
(serotype H-14).

conc. (ppm)	Averages of corrected Mortality percent		
	24 hrs.	48 hrs.	72 hrs
	$\bar{X} \pm SE$	$\bar{X} \pm SE$	$\bar{X} \pm SE$
0.5	100	100	100
0.15	90.26 \pm 0.59	93.88 \pm 0.56	98.02 \pm 0.78
0.1	80.96 \pm 0.78	90.52 \pm 0.67	94.16 \pm 0.30
0.05	52.10 \pm 0.65	70.18 \pm 0.68	83.20 \pm 0.61
0.01	16.64 \pm 0.97	28.72 \pm 0.95	47.88 \pm 0.04
0.005	5.18 \pm 0.67	12.88 \pm 0.92	27.62 \pm 0.63
0.001	0	10.88 \pm 0.63	14.3 \pm 0.33
0.0005	0	0	0
Lc50	0.037ppm	0.022ppm	0.013ppm
Lc90	0.15ppm	0.1ppm	0.064ppm

These values are the average of 11 tests. Each dilution has 6 replicates in which 10 larvae were treated.

The obtained data in table (1) indicate that mortality among treated larvae increased with the increase of the exposure time.

(P < 0.05)

Fig. (3)

Regression lines of mortality among the early
3rd instar larvae of Culex pipiens at 24, 48
and 72 hr. after treatment with different
concentrations of Bacillus thuringiensis H-14.

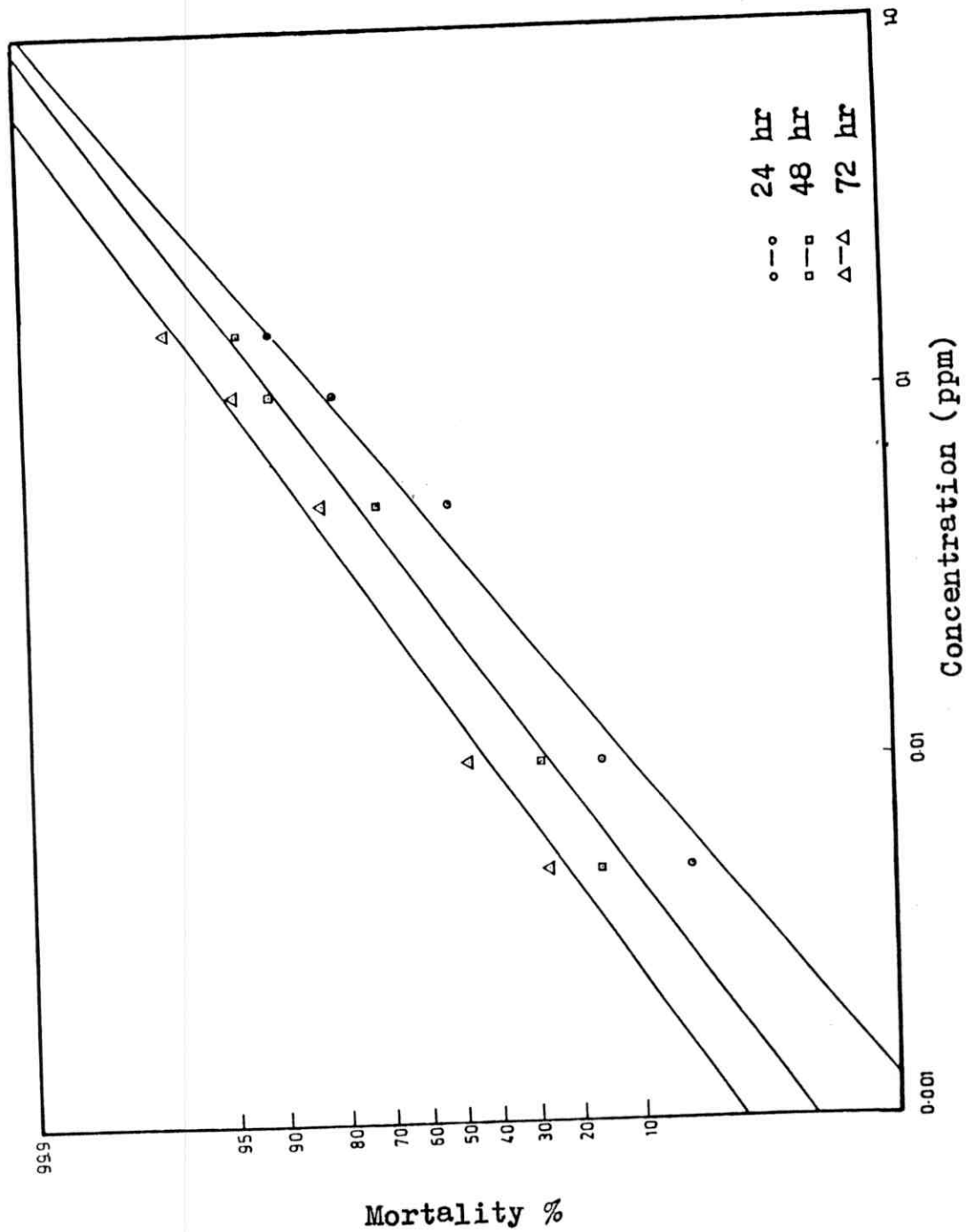


Fig. (3).

1.1.2. Bacillus sphaericus strain 1593-4 treatments:

Bacillus sphaericus was biologically assayed against the 3rd instar larvae. Mortality occurred after 24, 48 and 72 hours, post-treatments are presented in table (2) and graphically illustrated in Fig. (4).

The data in table (2) indicate the following:

- High concentrations evoked a rapid (24 hr) lethal response to the entire tested larvae subjected to that dose.
- The susceptibility of the treated larvae was significantly increased with the exposure time.

($P < 0.05$)

Table (2)

Sasceptibility of the early 3rd instar larvae of Culex pipiens to Bacillus sphaericus strain 1593-4.

Conc. (ppm)	Averages of corrected mortality percent		
	24hr.	48hr.	72hr.
	$\bar{X} \pm SE$	$\bar{X} \pm SE$	$\bar{X} \pm SE$
10	100	100	100
8	90.90 \pm 0.05	100	100
6	85.10 \pm 0.01	98.30 \pm 0.01	99.30 \pm 0.02
3	53.98 \pm 0.06	93.13 \pm 0.09	99.10 \pm 0.01
2	41.36 \pm 0.23	77.88 \pm 0.80	94.66 \pm 0.10
1.5	20.82 \pm 0.64	59.84 \pm 0.48	85.62 \pm 0.93
1	15.02 \pm 0.81	43.86 \pm 0.88	58.72 \pm 0.43
0.5	8.86 \pm 0.64	14.18 \pm 0.27	29.38 \pm 0.91
0.1	5.00 \pm 0.81	9.94 \pm 0.12	15.42 \pm 0.99
LC ₅₀	2.6 ppm	1.18 ppm	0.74 ppm
LC ₉₀	8 ppm	2.7 ppm	1.6 ppm

The mean is the average of 11 treatments each dilution has 6 replicates in which 10 larvae were treated.

Fig. (4)

Regression lines of mortality among the early 3rd instar larvae of Culex pipiens at 24, 48 and 72 hours, after treatment with different concentrations of Bacillus sphaericus 1593-4.

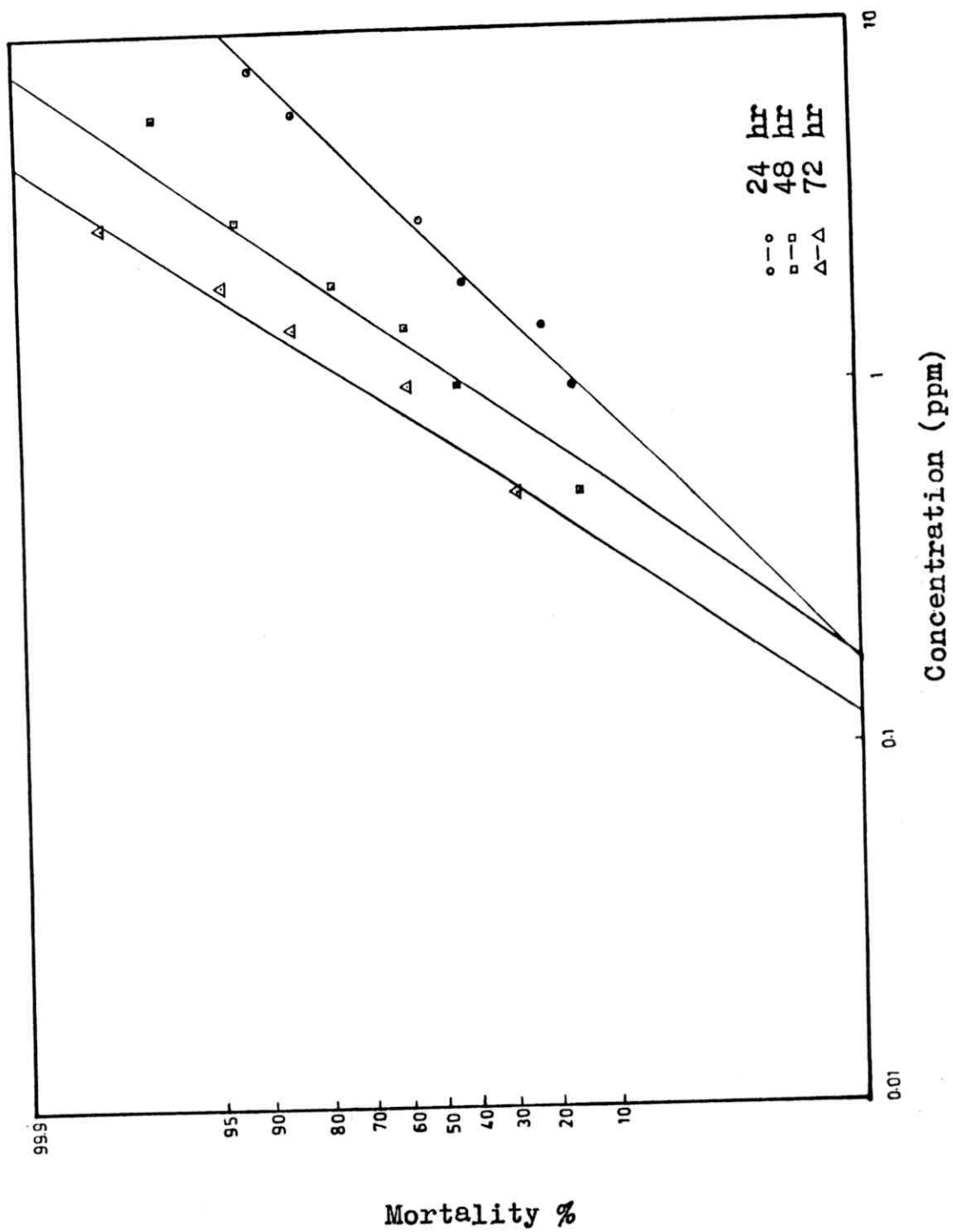


Fig. (4)

1.2. Chemical larvicide

1.2.1. Dieldrin treatments:

Dieldrin was assayed against the 3rd larval instar of Culex pipiens. Mortality resulted after 24, 48, and 72 hr., post-treatments are presented in table (3) and graphically illustrated in Fig. (5).

Table (3)

Susceptibility of the early 3rd instar larvae of Culex pipiens to dieldrin (chemical larvicide).

Conc. (ppm)	Averages of corrected mortality percent		
	24hrs.	48hrs.	72hrs.
	* $\bar{X} \pm SE$	$\bar{X} \pm SE$	$\bar{X} \pm SE$
0.04	100	100	100
0.03	91.18 \pm 0.01	96.38 \pm 0.33	98.60 \pm 0.57
0.02	70.92 \pm 0.02	83.56 \pm 0.68	93.24 \pm 0.66
0.015	62.66 \pm 0.12	76.20 \pm 0.15	84.40 \pm 0.48
0.01	30.74 \pm 0.70	39.82 \pm 0.28	58.86 \pm 0.91
0.005	7.70 \pm 0.11	14.04 \pm 0.37	24.48 \pm 0.42
0.001	0	10.1 \pm 0.55	12.7 \pm 0.42
0.0005	0	0	0
LC ₅₀	0.013	0.011	0.0086
LC ₉₀	0.027	0.022	0.018

* The mean is the average of 11 treatments each dilution has 6 replicates in which 10 larvae were treated.

The data in table (3) indicate that the susceptibility of the treated larvae was increased with the exposure time.
(P < 0.05)

Fig. (5)

Regression lines of mortality among the early 3rd instar larvae of Culex pipiens at 24, 48 and 72 hrs. after treatment with different concentrations of dieldrin chemical larvicide.

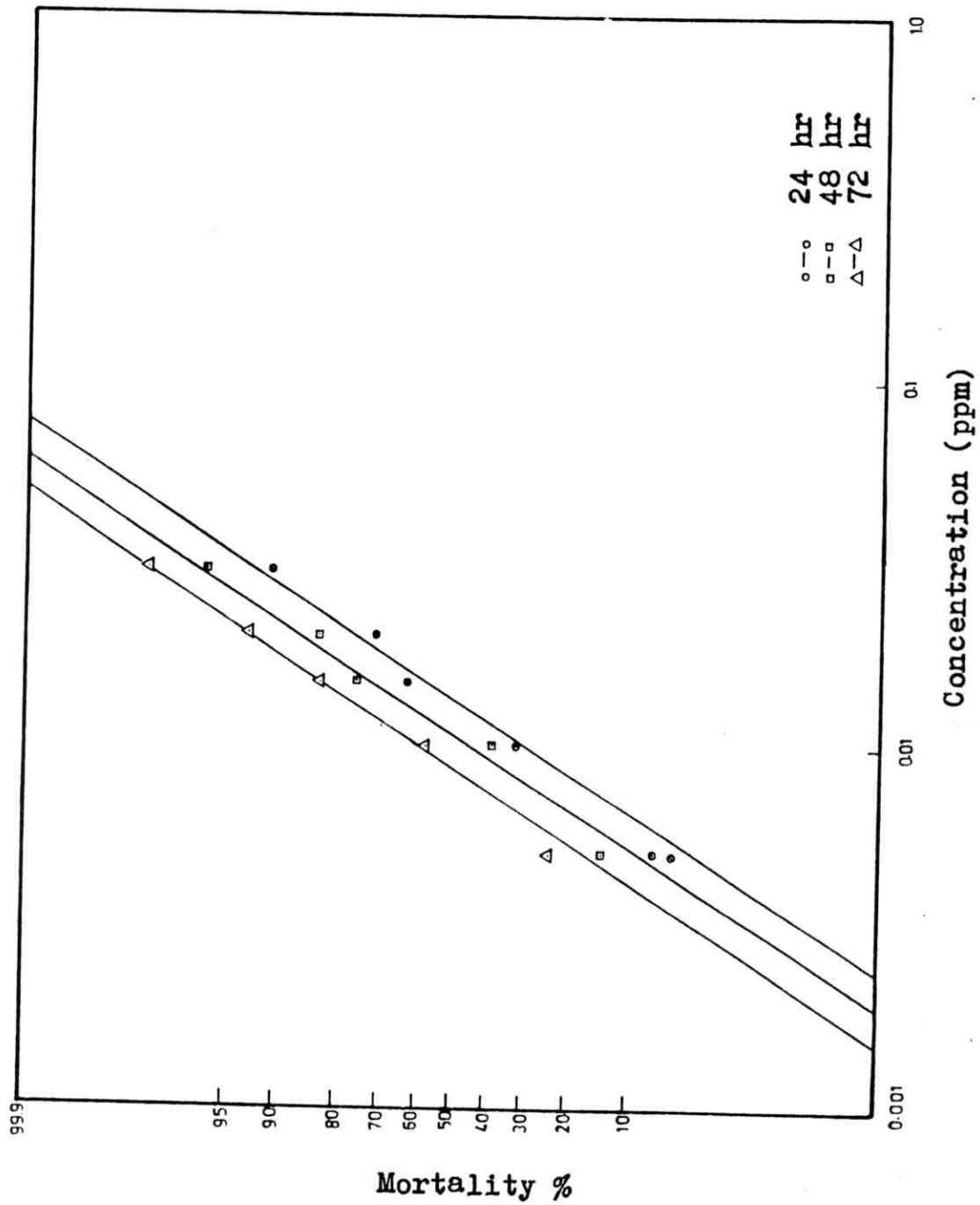


Fig. (5).

2. Comparative susceptibility of Culex pipiens larvae

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to the three tested larvicides:

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Based on the obtained results of the previous series of experiments, the susceptibility of the Culex pipiens 3rd instar larvae to the three larvicides was compared in order to evaluate their larvicidal activity. The tabulation of the estimated LC_{50} s and LC_{90} s of each of the three tested larvicides in table (4) indicate that the larvicidal activity was relatively very high due to larval exposure to dieldrin followed by Bacillus thuringiensis H-14 then Bacillus sphaericus 1593-4. However, the increase of mortality with exposure time was quite linear in case of both Bacillus thuringiensis H-14 and dieldrin while, the increase of mortality due to Bacillus sphaericus 1593-4 started to be significantly increased after 24 hr post-exposure.

($P < 0.05$)

Table (4)

Comparative susceptibility of the three used larvicides (Bacillus thuringiensis H-14, Bacillus sphaericus & dieldrin) during 24, 48 and 72 hrs. of Culex pipiens larval exposure.

Larvicide used	Lethal conc. in ppm	exposure time		
		24 hr.	48hr.	72hr.
<u>Bacillus</u> <u>thuringiensis</u>	LC ₅₀	0.037	0.022	0.013
	LC ₉₀	0.15	0.1	0.064
<u>Bacillus</u> <u>sphaericus</u>	LC ₅₀	2.6	1.18	0.74
	LC ₉₀	8	2.7	1.6
dieldrin	LC ₅₀	0.013	0.011	0.0086
	LC ₉₀	0.027	0.022	0.018

3. Combined Effect Of Mixed Formulations Of The
Chemical And Bacterial Larvicides.

3.1. Mixures of dieldrin and Bacillus thuringiensis
H-14.

A mixture of dieldrin and Bacillus thuringiensis H-14 was bioassayed against 3rd instar larvae of Culex pipiens at LC_{50} s. concentrations and the resulted larval mortalities after 24, 48, and 72 hr of exposure to the mixed suspensions are tabulated in table (5).

Data in table (5) indicate the following:

- The 3rd instar larvae of Culex pipiens were susceptible to the mixture of the Bacillus thuringiensis and dieldrin.
- Using (Bacillus thuringiensis and dieldrin) mixture resulted in a significant higher larval mortality than either of them when used separately.
- Larval mortality was directly correlated with exposure time. ($P < 0.05$)

Table (5)

Mortality of the early 3rd instar larvae of Culex pipiens treated with a mixture of dieldrin and B. thuringiensis after 24, 48 and 72 hr exposure periods.

Exposure	<u>B.t.i</u>		DLD		Averages of corrected mortality percent.
time	LC ₅₀ ppm	LC ₉₀ ppm	LC ₅₀ ppm	LC ₉₀ ppm	(<u>B.t.i</u> H-14 & DLD) mixture (0.037 ppm + 0.013 ppm)
					$\bar{X} \pm SE$
24 hr	0.037	0.15	0.013	0.027	70.80 \pm 0.28
48 hr	0.022	0.1	0.011	0.022	81.04 \pm 0.56
72 hr	0.013	0.064	0.0086	0.018	88.32 \pm 0.44

* The mean is the average of 6 treatments each of 6 replicates (10 larvae).

3.2. Mixures of dieldrin and Bacillus sphaericus
1593-4.

A mixture of dieldrin and Bacillus sphaericus 1593-4 was bioassayed against 3rd instar larvae of Culex pipiens at LC_{50} s concentrations and the resulted larval mortalities after 24, 48, and 72hr of exposure to the mixed suspension are tabulated in table (6).

Data in table (6) indicate the following:

- The early 3rd instar larvae of Culex pipiens are susceptible to the mixture of dieldrin and Bacillus sphaericus.
- Significant larval mortality occur in using the mixture of (dieldrin and Bacillus sphaericus) than if either of them was used alone.
- Increasing the exposure time result in significant mortality to larvae exposed. ($P < 0.05$).

Table (6)

Mortality of the early 3rd instar larvae of Culex pipiens treated with a mixed formulations of dieldrin and Bacillus sphaericus 1593-4 after 24,48 and 72 hr exposure period.

Exposure time	<u>B. spha.</u>		DLD		Averages of corrected mortality percent.
	LC ₅₀ ppm	LC ₉₀ ppm	LC ₅₀ ppm	LC ₉₀ ppm	(<u>B. spha.</u> & DLD) mixture (2.6 ppm + 0.013 ppm)
					$\bar{X} \pm SE$
24 hr	2.6	8	0.013	0.027	69.50 \pm 0.83
48 hr	1.18	2.7	0.011	0.022	85.84 \pm 0.97
72 hr	0.74	1.6	0.0086	0.018	94.30 \pm 0.78

* The mean is the average of 6 treatments each has 6 replicates in which 10 larvae were treated.

4. The Joint Action Between The Bacterical Larvicides.

4.1. Mixures of *Bacillus thuringiensis* H-14 and
Bacillus sphaericus 1593-4.

A mixture of *Bacillus thuringiensis* H-14 and *Bacillus sphaericus* was bioassayed against 3rd instar larvae of *Culex pipiens* at the LC_{50} s levels and the resulted larval mortalities after 24, 48, and 72hr of exposure to the mixed suspension are tabulated in table (7).

Data in table (7) indicate the following:

- *Culex pipiens* larvae are quite susceptible to the bacterial mixture (*Bacillus thuringiensis* H-14 and *Bacillus sphaericus* 1593-4).
- The use of the bacterial mixture result in significant larval mortality than if either of them was used alone.
- Increasing the exposure time result in significant larval mortality to larvae exposed.
($P < 0.05$).

Table (7)

Percentage mortality of the early 3rd instar larvae of Culex pipiens treated with a mixed formulations of Bacillus thuringiensis H-14 and Bacillus sphaericus 1593-4 at 24,48 and 72 hr exposure period.

Exposure time	<u>B.t.i</u>		<u>B.spha.</u>		Averages of corrected mortality percent.
	LC ₅₀ ppm	LC ₉₀ ppm	LC ₅₀ ppm	LC ₉₀ ppm	(<u>B.t.i</u> & <u>B.spha.</u> (0.037ppm+ 2.6ppm) mixture
					$\bar{X} \pm SE$
24 hr	0.037	0.15	2.6	8	78.82 \pm 0.08
48 hr	0.022	0.1	1.18	2.7	96.91 \pm 0.34
72 hr	0.013	0.064	0.74	1.6	100 \pm 0

* The mean is the average of 6 treatments each has 6 replicates in which 10 larvae were treated.

5. Comparative Susceptibility Of Culex pipiens
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Larvae To The Larvacides Mixtures.
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The larval mortality resulted from treating the 3rd instar larvae of Culex pipiens with the following mixture of:

- 1) (Bacterial + chemical) larvicides
 - 1.1. (Bacillus thuringiensis + dieldrin) mixture.
 - 1.2. (Bacillus sphaericus + dieldrin) mixture.
- 2) Bacterial mixture

(Bacillus thuringiensis + Bacillus sphaericus) mixture were represented in table (8) as follows.

The data in Table (8) indicat the following:

- There is a significant increase in larval mortality occur from the use of larvicides mixture ($P < 0.05$)
- Comparison between the combined effect of the mixture of (Bacillus thuringiensis + dieldrin) and the mixture of (Bacillus sphaericus + dieldrin) indicate that there is no significance difference between the mortality percent occur to 3rd instar Culex pipiens larvae at 24 and 48hr exposure time, but at 72hr exposure period there is a significance difference show that the

Table (8)

Percentage mortality of the early 3rd instar larvae of Culex pipiens treated with mixtures of (Bacillus thuringiensis + dieldrin), (Bacillus sphaericus + dieldrin) or (Bacillus thuringiensis + Bacillus sphaericus)

Combined mixture of LC ₅₀ s of the tested larvicides	Averages of corrected mortality percent		
	exposure time		
	24 hr	48 hr	72 hr
	$\bar{X} \pm SE$	$\bar{X} \pm SE$	$\bar{X} \pm SE$
(<u>B.T.I</u> H-14+ DLD) 0.037 ppm + 0.013ppm	70.80 \pm 0.28	81.04 \pm 0.56	88.32 \pm 0.44
(<u>B.spha.</u> + DLD) 2.6 ppm + 0.013 ppm	69.50 \pm 0.83	85.84 \pm 0.97	94.30 \pm 0.78
(<u>B.T.I</u> H-14+ <u>B.spha.</u>) 0.037ppm + 2.6ppm	78.82 \pm 0.08	96.91 \pm 0.34	100

(Bacillus sphaericus +dieltrin) mixture is effective.
($P < 0.05$)

- Comparison between the combined effect of the bacterial mixture (Bacillus thuringiensis + Bacillus sphaericus) and (Bacillus thuringiensis +dieltrin) mixture indicate that the first mixture have a significant effect than the second mixture.
($P < 0.05$)

- The bacterial mixture (Bacillus thuringiensis + Bacillus sphaericus) have highly significant effect than (Bacillus sphaericus + dieltrin) mixture.
($P < 0.05$)

6. Effect Of Using The Bacterial And Chemical Larvicides
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On Culex pipiens Larvae In **Sequence**;
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The susceptibility of the 3rd instar larvae of Culex pipiens to the toxic effect of the entomopathogenic bacteria, Bacillus thuringiensis H-14 or Bacillus sphaericus when pre-treating the larvae with it for 24 hr and then the other bacteria or dieldrin was added, were evaluated through carrying out a series of laboratory experiments.

The results obtained from these experiments were as follows:

- 6.1. Larvae pre-treated with Bacillus thuringiensis H-14
for 24 hr and then Bacillus sphaericus or dieldrin
was added.

Results of pre-treating 3rd instar larvae of Culex pipiens with Bacillus thuringiensis H-14 for 24 hr then adding Bacillus sphaericus or dieldrin larvicides were tabulated in table (9) as the following.

Table (9)

Percentage mortality of the early 3rd instar larvae of Culex pipiens pre-treated with B. thuringiensis H-14
 LC_{50} (0.037 for 24 hr to Bacillus sphaericus LC_{50} (2.6 ppm) or dieldrin LC_{50} (0.013 ppm)

Post-Exposure time	<u>B. spha.</u>		DLD		Averages of corrected mortality percent.	
	LC_{50} ppm	LC_{90} ppm	LC_{50} ppm	LC_{90} ppm	Larvae pre-treated with <u>B.t.i.</u> for the first 24hr with LC_{50} conc. = 0.037ppm	
24 hr	2.6	8	0.013	0.027	Larvae post-mixed treatment with <u>B. spha.</u>	Larvae post-mixed treatment with dieldrin.
					$\bar{X} \pm SE$	$\bar{X} \pm SE$
					77.30 \pm 0.05	88.18 \pm 0.39
48 hr	1.18	2.7	0.011	0.022	99.99 \pm 0.01	96.12 \pm 0.98

Data in table (9) indicate the following:

- Adding Bacillus sphaericus to larvae pre-treated with Bacillus thuringiensis for 24 hr cause significant mortality than if Bacillus sphaericus was used alone.
- Pre-treating the Culex pipiens 3rd instar larvae with Bacillus thuringiensis for 24 hr then dieldrin was added result in a significant larval mortality than if dieldrin was used alone.
- Pre-treating the larvae with Bacillus thuringiensis H-14 for 24 hr and then dieldrin was added have a significant increase in larval mortality in the first 24 hr after postmixed treatment, but with increasing exposure time no significant mortality occur than adding Bacillus sphaericus.

6.2. Larvae pre-treated with Bacillus sphaericus
1593-4 for 24 hr and then Bacillus thuringiensis
or dieldrin was added.

Treating the 3rd instar Culex
pipiens larvae with Bacillus sphaericus for 24 hr
with the L_{50} Conc= 2.6 ppm then Bacillus
thuringiensis H-14 or dieldrin larvicides was
added inorder to evaluate its effectivness were
represented in table (10) as the following:

Table (10)

Percentage mortality of the early 3rd instar larvae of Culex pipiens pre-treated with Bacillus sphaericus 1593-4 LC₅₀ (2.6 ppm) for 24 hr to Bacillus thuringiensis H-14 LC₅₀ (0.037 ppm) or dieldrin LC₅₀ (0.013 ppm).

Post-Exposure time	<u>B.t.i.</u>		DLD		Averages of corrected mortality percent	
	LC ₅₀ ppm	LC ₉₀ ppm	LC ₅₀ ppm	LC ₉₀ ppm	Larvae pre-treated with <u>B. spha.</u> for the first 24 hr with LC ₅₀ conc. = 2.6 ppm	
24 hr	0.037	0.15	0.013	0.027	Larvae post-mixed treatment with <u>B.t.i.</u> H-14	Larvae post-mixed treatment with dieldrin
					$\bar{X} \pm SE$	$\bar{X} \pm SE$
					90.02 \pm 0.35	85.95 \pm 0.21
48 hr	0.022	0.1	0.011	0.022	100	99.01 \pm 0.30

Data in table (10) indicate the following

- Pretreating the Culex pipiens larvae with Bacillus sphaericus for 24 hr then Bacillus thuringiensis was added cause a significant increase in larval mortality than if Bacillus thuringiensis was used alone.
- Adding dieldrin to larvae pre-treated with Bacillus sphaericus cause significant larval mortality than if dieldrin was used alone.
- Adding Bacillus thuringiensis H-14 to larvae pre-treated with Bacillus sphaericus for 24 hr cause a significant larval mortality than if dieldrin was added ($P < 0.05$).

7. Effect Of Pre-treating Culex pipiens Larvae With
=====
Dieldrin For 24 hr Then Bacterial Larvicides
=====
Was Added.
=====

Finding the potential efficacy of dieldrin larvicide when the 3rd instar of Culex pipiens larvae were pre-treated with it for 24 hr with the LC_{50} concentration= 0.013 ppm and then Bacillus thuringiensis or Bacillus sphaericus was added; were done through carrying out a series of laboratory experiments. The result was represented in table (11) as the following:

Table (11)

Percentage mortality of early 3rd instar larvae of Culex pipiens pre-treated with dieldrin LC₅₀ (0.013 ppm) for 24 hr to Bacillus thuringiensis H-14 LC₅₀ (0.037 ppm) or Bacillus sphaericus 1593- 4 LC₅₀ (2.6 ppm).

Post-exposure time	<u>Bti</u> H-14		<u>B. sphaericus</u>		Averages of corrected mortality percent	
	LC ₅₀ ppm	LC ₉₀ ppm	LC ₅₀ ppm	LC ₉₀ ppm	Larvae pre-treatment with dieldrin for the first 24hr with LC ₅₀ conc= 0.013 ppm	
24 hr	0.037	0.15	2.6	8	Larvae post-mixed treatment with <u>Bti</u> H-14.	Larvae post-mixed treatment with <u>B. sphaericus</u> .
					$\bar{x} \pm SE$	$\bar{x} \pm SE$
					92.11 \pm 0.10	88.99 \pm 0.31
48 hr	0.022	0.1	1.18	2.7	97.26 \pm 0.04	98.87 \pm 0.40

Data in table (11) indicate the following:

- Pretreating the Culex pipiens larvae for 24 hr with dieldrin then Bacillus thuringiensis was added causes significant increase in larval mortality than if the Bacillus thuringiensis was used alone.
- Adding the Bacillus sphaericus to larvae pre-treated with dieldrin for 24 hr causes significant increase in larval mortality than if Bacillus sphaericus was used alone.
- There was no significance difference in larval mortality if the Bacillus thuringiensis H-14 or Bacillus sphaericus was added to larvae pretreated with dieldrin for 24 hr.

8. Comparative susceptibility of Culex pipiens larvae
=====
to the larvicide mixtures when used in sequence :
=====

The results of pre-treating the Culex pipiens larvae for 24 hr with bacterial larvicides and then the other chemical or bacterial larvicides was added and vice-versa was represented in table (12) as follows .

Table (12)

Comparative susceptibility of early 3rd Culex pipiens larvae pre-treated with dieldrin, Bacillus thuringiensis H-14 or Bacillus sphaericus 1593-4 for 24 hr. then the other larvicides was added.

Larvae pre-treated for 24 hr with	The added Larvicide	Averages of corrected mortality percent	
		Exposure time	
		24 hr	48 hr
<u>B. thuringiensis</u> serotype H-14	<u>B. sphaericus</u> dieldrin	$\bar{x} \pm SE$	$\bar{x} \pm SE$
		77.30 \pm 0.05	99.99 \pm 0.01
		88.18 \pm 0.39	96.12 \pm 0.98
<u>B. sphaericus</u> 1593-4	<u>Bti</u> H-14 dieldrin	90.02 \pm 0.35	100
		85.95 \pm 0.21	99.01 \pm 0.30
dieldrin	<u>Bti</u> H-14 <u>B. sphaericus</u>	92.11 \pm 0.10	97.26 \pm 0.04
		88.99 \pm 0.31	98.87 \pm 0.40

Data in table (12) reveals the following:

- There is no significance differences occur to larval mortility rates when the Culex pipiens larvae pretreated for 24 hr with Bacillus thuringiensis H-14 or Bacillus sphaericus 1593-4 and then dieldrin was added
($P < 0.05$)
- Mortility persent occur as a results of pretreating the larvae for 24 hr with Bacillus sphaericus and then Bacillus thuringiensis was added is significant; than that occur from pretreated with Bacillus thuringiensis for 24 hr and then Bacillus sphaericus was added.
($P < 0.05$)
- There was no significance difference between larval mortility occured from pretreating Culex pipiens larvae with dieldrin for 24 hr then Bacillus sphaericus was added and pretreating the larvae with Bacillus sphaericus for 24 hr then dieldrin was added.
($P < 0.05$)
- No significance difference of the larval mortility occur between pretreating the larvae with dieldrin for 24 hr then Bacillus thuringiensis was added and pretreating the larvae with Bacillus thuringiensis for 24 hr then dieldrin was added.
($P < 0.05$)

9. Histopathological Studies:

The pathological response of the different body organs of Culex pipiens larvae due to the toxic action of the chemical larvicide (dieldrin) and two bacterial larvicides namely; Bacillus thuringiensis H-14 and Bacillus sphaericus 1593-4 was investigated.

The interaction between bacterial and chemical mosquito larvicides was also studied, histopathologically. Microscopic examination of the prepared sections of treated larvae reveals the followings:-

9.1) Bacillus thuringiensis H-14 treated larvae.

The toxic effect on the larval different tissue cells due to Bacillus thuringiensis H-14 infection was demonstrated in the Figures as the following:

1- Alimentary canal

The pathological action on the epithelial mid-gut cells first appeared as an enlargement of the nucleus followed by the darkening of the cytoplasm (Fig. 10 a) in comparison with the normal (Fig. 6a and 6b).

The cell size enlarge and the cytoplasm became vacuolated then the cell wall towards the gut lumen protrudes to form the characteristic balloon-shaped cell.

At this stage the cellular linear layer is partly

detached from the basement membrane (Figs. 10a, 10b and 11). Due to the increase of cell size, the cell membrane became ruptured, liberating the cytoplasm or part of it to be discharged in the gut lumen (Figs. 11 and 14). Goblet cells are also found to be responded to the toxic action of the ingested bacteria where the cytoplasm is changed to black colour (Fig. 10b).

In moribund larvae the peritrophic membrane seem to be not damaged by the treatment i.e. at the early stage of infection (Fig. 13). The regenerative conical-shaped cells are also affected (Fig. 10a). On higher magnification (Figs. 15 and 16) in advanced treated larvae most cells appeared hypertrophied, sloughed and completely damaged. The examination of sections of either the fore- or hind-gut regions, the epithelium at these regions were found not to be influenced by the toxic action of Bacillus thuringiensis H-14. However; the section representing the gastric caeca showed pathological pictures almost similar to that observed at the mid-gut region (Fig. 14). The columnar gland cells of the gastric caeca show hypertrophy, some cells are completely lysed and the cytoplasmic vacuolation is seen in the hypertrophied cells.

2- Muscle cells

The muscle layers surrounding the gut are detached from the epithelial gut cells. However, the muscle cells appear normal (Fig. 12)

3- Fat tissues

The fat body cells appeared destroyed (Fig. 13 & 15); they became highly vacuolated and underwent necrosis some of the ribbon-like tissues were detached from one another.

4- Malpighian tubules

The Malpighian tubules lost their striated brush borders and the cells became larger in size than the normal which appeared to rest on opposite sides thus this enlargement cause the narrowing of the tube lumen (Figs. 17&18).

5- Nerve cord

The examination of the prepared sections clears that the nerve cells and or ganglia are not affected by the bacterial infection.

6- Tracheal tubes

Tracheal tubes appeared normal, however,

the cells forming the tracheal matrix showed the characteristic symptoms of bacterial infection (Fig. 19).

9.2. Bacillus sphaericus 1593-4 treated larvae.

Sections in Culex pipiens larvae that were treated with Bacillus sphaericus 1593-4 showed the following features.

1- Alimentary canal

Preliminary screening of the longitudinal sections of the treated larvae for pathological changes in comparison with those from normal larvae (Fig. 6a) indicate that the epithelial lining of the alimentary canal seem to exhibit extensive cellular damage with distorted nuclei (Fig. 20).

In advanced dark brown cadavers the epithelial lining cells appeared to be enlarged not towards the gut lumen but it seem to be elongated along its own border. The nuclei seem to be enlarged, evacuated, at late stage of infection it could be seen in close contact with each other (Fig. 21).

The posterior mid-gut zone exhibited progressive degeneration. Cells became highly vacuolated, pyknotic and finally sloughed off, which allowed the bacteria access to the hemocoel. The gastric caeca cells was also affected (Fig. 21).

2- Muscle cells

The muscles appeared detached from their own bundle shaped structure.

3- Fat tissues

The fat cells were also affected .

4- Malpighian tubules

The Malpighian tubules cells also show the cellular hypertrophy (Fig. 21).

5- Nerve cord

The nerve cord element seem to be somewhat not affected by the infection.

6- Tracheal tubes

The tracheal matrix cells were affected as shown in (Fig. 22).

9.3. Dieldrin treated larvae.

Section in larvae that were treated with the chemical larvicide dieldrin showed to be affected in many features as the following:

1- Alimentary canal

The lining epithelial cells of the alimentary canal (Fig. 23) were affected, the cytoplasm of

these cells showed signs of a cloudy swelling. The nuclei appeared pyknotic. The basal regenerative cells "nidi" still present among the lining epithelial cells. The gastric caeca cells were also affected.

2- Muscle cells

The muscles were affected by dieldrin treatment. The myofibrils shrunk and were grouped into masses separated by vacuoles and fissures (Fig. 24).

3- Fat tissues

The fat cells seemed to be highly vacuolated and underwent necrosis (Fig. 25). Some of the ribbon like tissues were detached from one another.

4- Malpighian tubules

The Malpighian tubules cells underwent degeneration (Fig. 26). Signs of necrosis and degeneration were evident in the nuclei and in the cytoplasm of these cells.

5- Nerve cord

The element of the nerve cord (Fig. 24) became very loosely packed. Some nerve cells lost the characteristic granular appearance in comparison with that appeared in untreated larvae (Fig. 8).

Others had highly vacuolated cytoplasm. The neuropile mass underwent a reduction in size. The cortical layer underwent degeneration.

6- Tracheal tubes

The tracheal tubes with its tracheal matrix cells (Fig. 25) were also, show the cellular hypertrophy.

9.4. Larvae treated with (dieldrin + Bacillus thuringiensis) mixture.

The longitudinal sections in (Fig. 27) and (Fig. 28) in a larva treated with (dieldrin + Bacillus thuringiensis) mixture revealed the destruction occur to the epithelial cells of the alimentary canal.

In (Fig 29) and (Fig. 30) the gut epithelia appeared swollen with vacuolated cytoplasm and pyknotic nuclei. The cells appeared separated at the base from each other. Also the regenerative cells which rest inbetween the epithelial cells became swollen. In a transverse section (Fig. 31) the fore gut epithelial cells , show the sign of damage.

In the mid-gut zone (Fig. 32) the epithelial cells appeared to be extensively damaged by such mixture. Some cells became swollen with an

empty apical portion. Others its membrane was ruptured releasing its cytoplasm inside the gut lumen. The peritrophic membrane was destroyed as a result of such treatment.

The gastric caeca appeared to be affected also, the muscle bundle and fat tissue became very loosely packed (Fig. 32)

A striking feature appeared in a longitudinal section with much higher magnification (Fig. 33) showing the epithelial cell with the typical balloon-shaped outgrowth towards the gut lumen. This sign was a characteristic symptom for the larvae treated with the Bacillus thuringiensis H-14, however it appeared with larvae treated with the mixture of this bacteria and dieldrin.

9.5. Larvae treated with (dieldrin + Bacillus sphaericus) mixture.

Treated larvae section in (Fig. 34) and (Fig. 35) with the (dieldrin + Bacillus sphaericus) mixture show that the gut epithelial cells affected by this mixture. The cytoplasm was highly vacuolated. The muscles appeared hypertrophied.

The mid-gut epithelial cell became swollen. The peritrophic membrane was destroyed.

The posterior part of mid-gut epithelial cell was extensively affected. Cellular hypertrophy appeared clearly. In (Fig. 36) the cytoplasm was released inside the gut lumen as a result of cell membrane rupture.

The muscle bundles surrounding the gut epithelial cell (Fig. 37) were affected.

In the proventriculus zone (Fig. 38) the epithelial cell was affected by the (dieldrin + Bacillus sphaericus) mixture. It became a separated spindle shaped cell. The cytoplasm was vacuolated, as well as, the nucleus of the cells.

A transverse section (Fig. 39) show that the tracheal tube became flattened and the tracheal matrix was also destroyed.

In a post-mortum larva (Fig. 40) the gut epithelial cell was highly destroyed. The gastric caeca was affected, its nucleus became enlarged

9.6. Larvae treated with (Bacillus thuringiensis + Bacillus sphaericus) mixture.

Examination of the sectioned material indicated the following:

The photographs in (Fig. 41, 42, 43, and 44) demonstrate a general view of longitudinal section

of treated larvae with the bacterial (Bacillus thuringiensis + Bacillus sphaericus) mixture. The epithelial mid-gut layer showed the monolayer structure with the indication of cellular hypertrophy. The cells are swollen with sloughing and vacuolated cytoplasm. Some apical zone of the epithelial cells appeared empty with a characteristic balloon-shaped out growths. The nuclei are enlarged showing pyknotic characteristics, some are completely vacuolated.

The peritrophic membrane is also affected by such treatment with apparent signs of damage.

In advanced stage in treated larvae (Fig. 45) the cell membrane is ruptured releasing the cytoplasm inside the gut lumen. The cytoplasmic vacuoles could be seen clearly. Some cells (Fig. 46) seem separate spindle shaped cells.

The muscle layers surrounding the gut are also affected by the bacterial mixture (Fig. 47).

Fat bodies and Malpighian tubules (Fig. 48) are affected. The Malpighian tubules cells are enlarged, others with a rupture membrane. The Malpighian tubules characteristic lumen result from resting of cells on opposite sides towards each other could not be seen. The nerve cord seems to be not affected by the bacterial mixture.

Fig. (6a) A longitudinal section of a non treated larva at the mid-gut region.

MB: Muscle bundle.

BM: Basement membrane.

EC: Epithelium cell.

GL: Gut lumen.

PM: **Peritrophic** membrane.

(X = 200)

Fig. (6b) : The same as Fig.(6a) at higher magnification.

EC: Epithelium cell.

GC: Gut contents.

MB: Muscle bundle.

(X = 400)

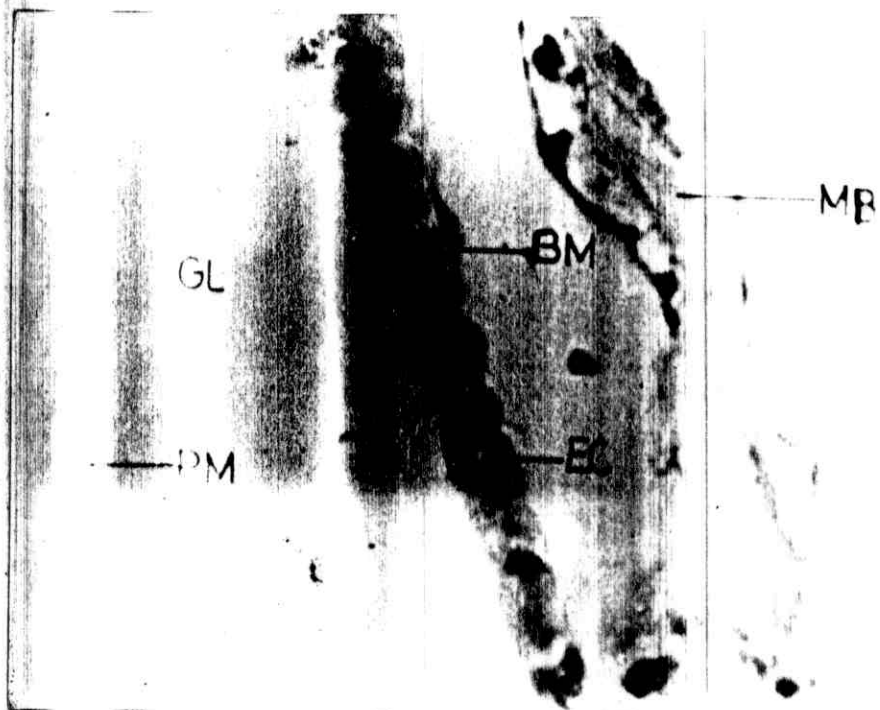


Fig. (6 a)

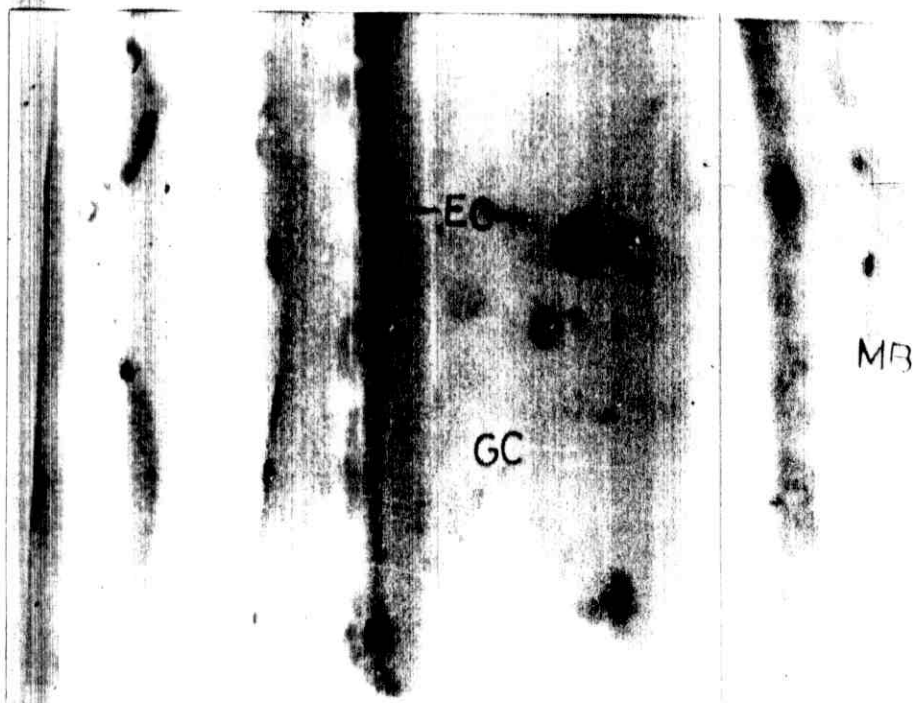


Fig. (6 b)

Fig. (7a): A transverse section in a non treated larva at the mid-gut region.

EC: Epithelium cell.
GC: Gut contents.
PM: Peritrophic membrane.
TT: Tracheal tube.
MB: Muscle bundle.

(X = 100)

Fig. (7b): The same as in Fig. (7a) at higher magnification.

GC: Gut contents.
PM: Peritrophic membrane.
EC: Epithelium cell.

(X = 400)

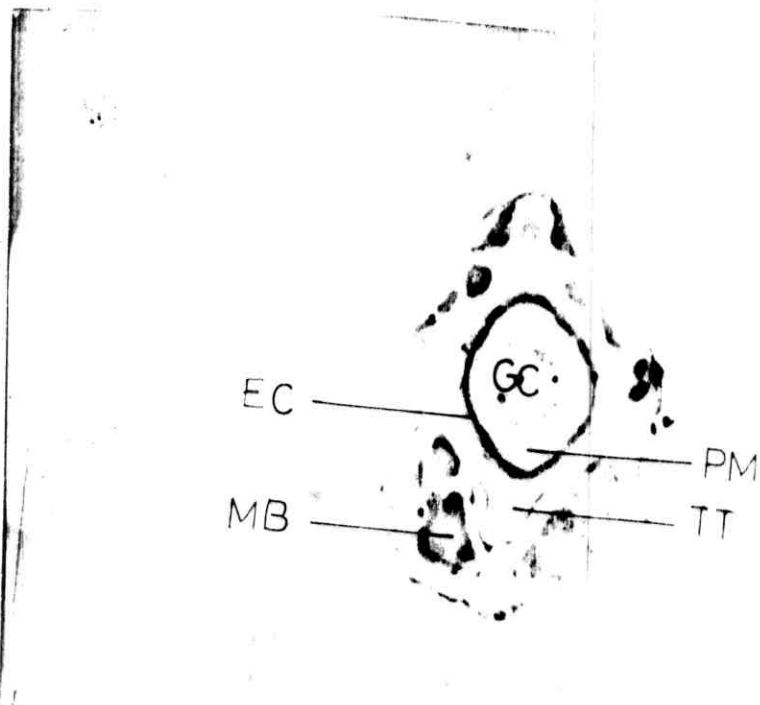


Fig. (7 a)



Fig. (7 b)

Fig. (8) : A transverse section through the thoracic region of a non treated larva.

CL: Cortical Layer.
NM: Neuropile mass.
EC: Epithelium cell.
GL: Gut lumen.

(X = 400)

Fig. (9): A transverse section of a non treated larva

TT: Tracheal tube.

(X = 400)

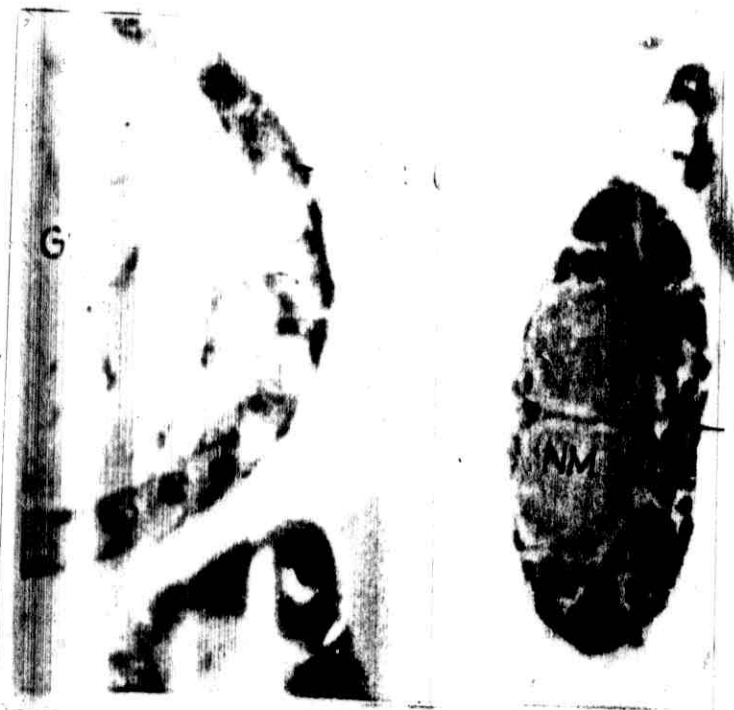


Fig. (8)



Fig. (9)

Fig. (10a): A longitudinal section in an infected larva
(moribund) treated with Bacillus thuringiensis
serotype H-14. Mid-gut region.

BSEC: **Balloon-** shaped epithelial cell.

PM: **P**eritrophic membrane.

GC: Gut contents.

SEC: Swallen epithelium cell.

NH: Nucleus hypertrophy.

SRC: Swallen regenerative cell.

(X = 400)

Fig. (10b): The same treated larva (10a)

NH: Nucleus hypertrophy.

SEC: Swallen epithelium cell.

GL: Gut lumen.

GC: Goublet cell.

(X = 400)

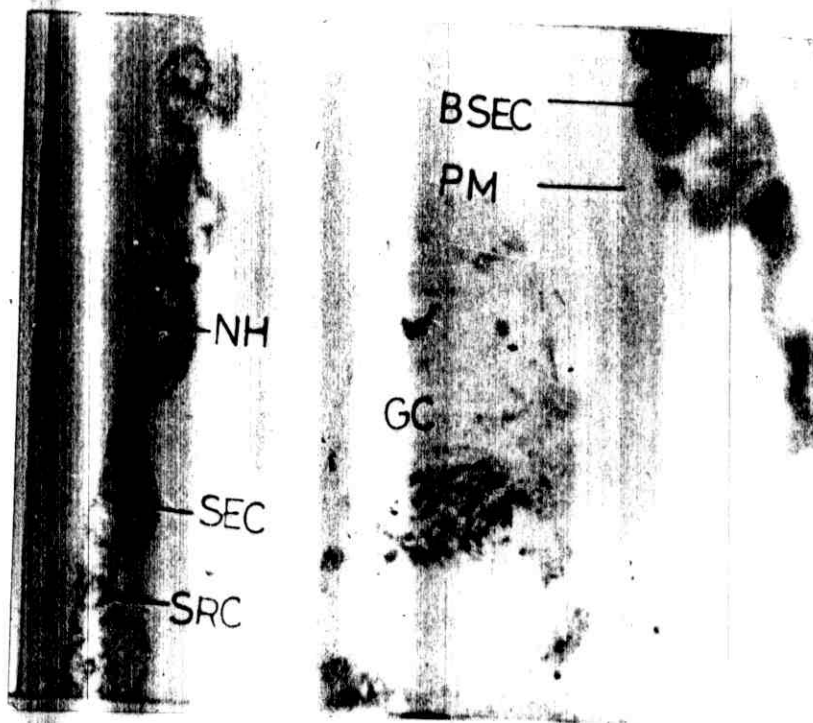


Fig. (10 a)



Fig. (10 b)

Fig. (11) : Photomicrograph of a longitudinal section at the posterior part of a gut of a larva infected with B. thuringiensis serotype H-14.

DPM : Destroyed peritrophic membrane.
GC : Gut contents.
EB : Empty apical portion.
FB : Fat body.

(X = 400)

Fig. (12) : Alongitudinal section of an infected larva with B. thuringiensis serotype H-14 (post-mortum).

RCM : Ruptured cell membrane.
VN : **Vacuolated** nucleus.
MB : muscle bundle.

(X = 400)

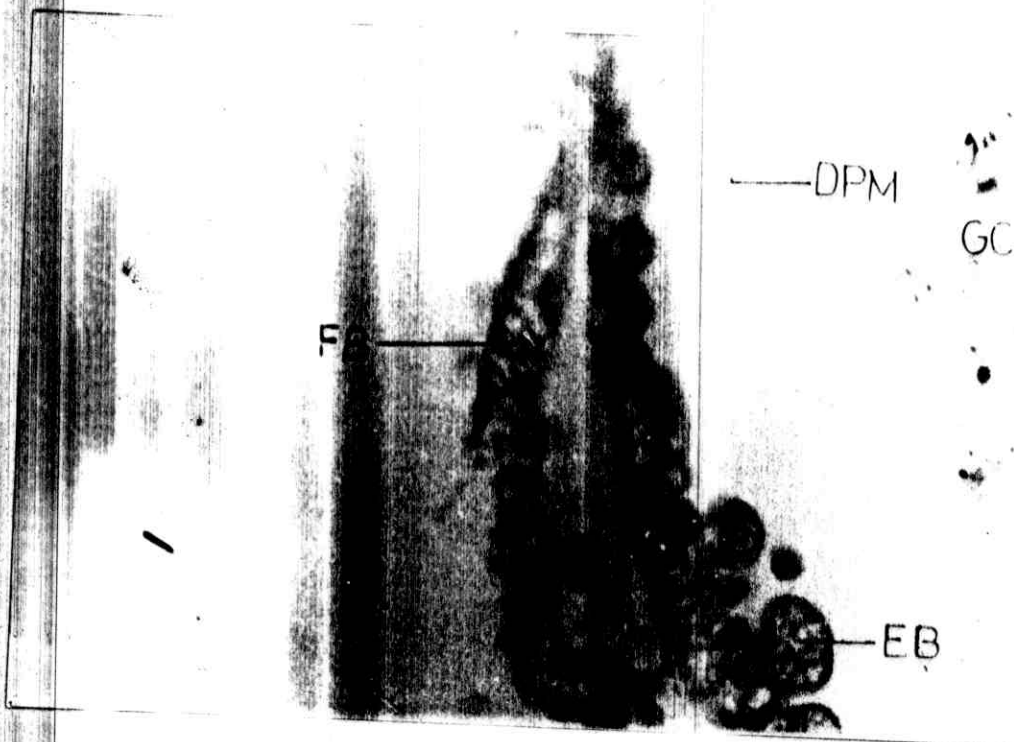


Fig. (11)

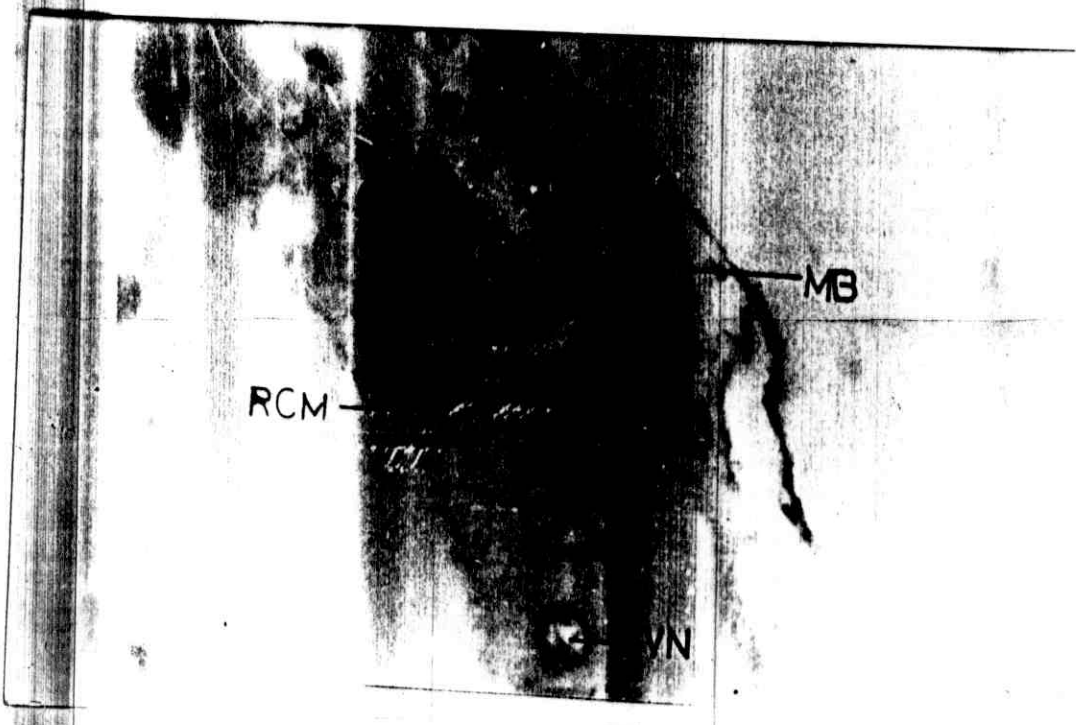


Fig. (12)

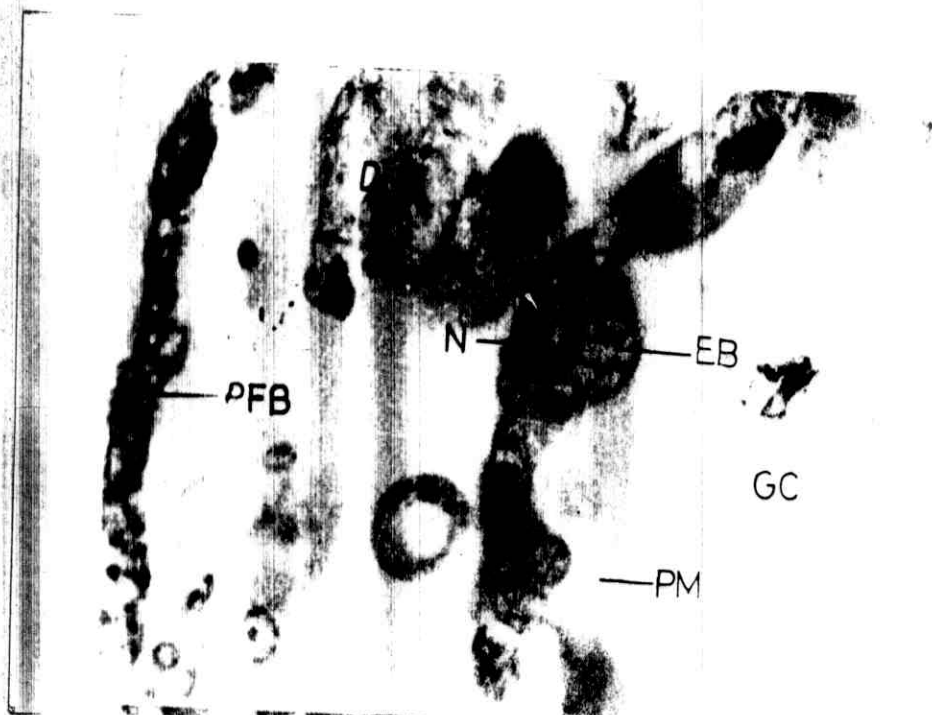


Fig. (13)

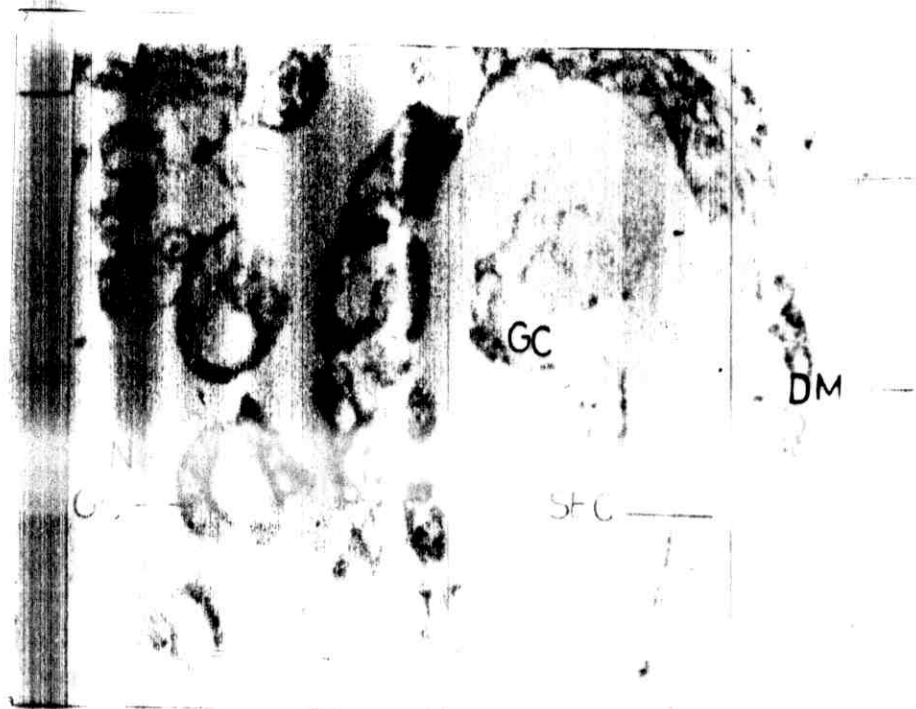


Fig. (14)

Fig. (15) : ~~Another transverse~~ section of a larva treated with B. thuringiensis (post-mortum).

GL : Gut lumen.
DEC : **Destroyed** epithelium cell.
DMB : **Destroyed** muscle bundle.
NH : Nucleus Hypertrophy.
DFT : **Destroyed** fat tissue.

(X = 400)

Fig. (16): A transverse section of a larva treated with B. thuringiensis (post-mortum).

CH : Cell hypertrophy.
ROC : Release of cytoplasm.
GC : Gut contents.
NH : Nucleus hypertrophy.
DEC : **Destroyed** epithelium cell.
DMB : **Destroyed** muscle bundle.
DFT : **Destroyed** fat tissue.

(X = 400)

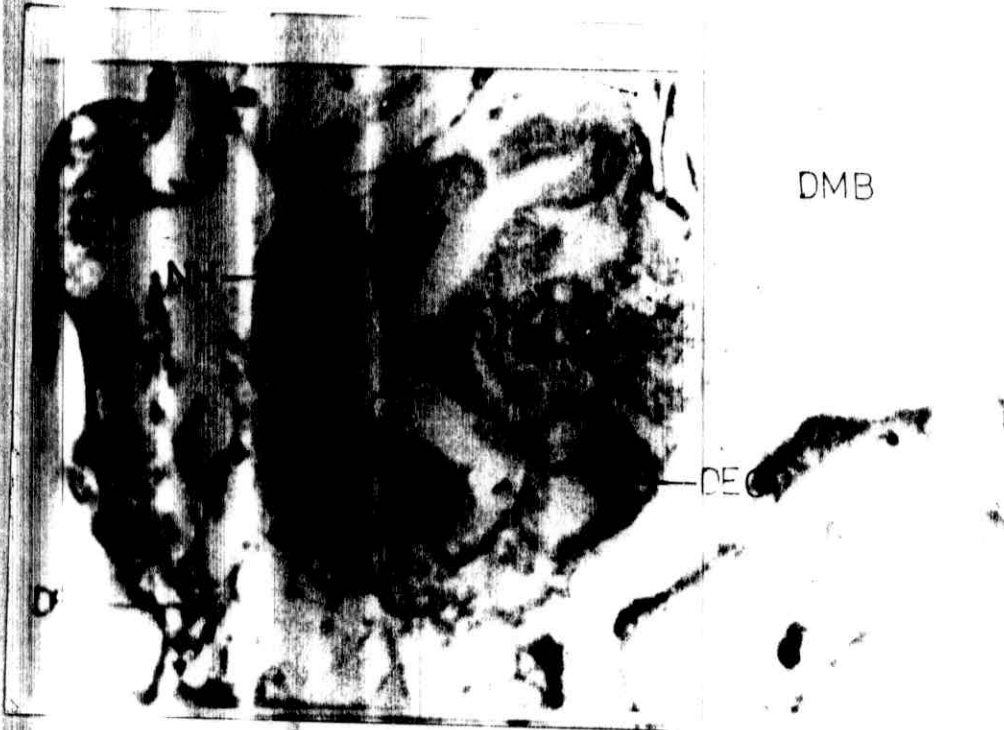


Fig. (15)



Fig. (16)

Fig. (17) : A longitudinal section of a larva treated with B. thuringiensis (moribund). Showing the Malpighian tubules in early stage of infection.

DMT : Destroyed Malpighian tubules.
NH : Nucleus hypertrophy.
DMB : Destroyed muscle bundle .
DPFB : Destroyed partial fat body.

(X = 400)

Fig. (18): Another longitudinal section of a larva treated with B. thuringiensis (post-mortum). Showing the disappearance of the Malpighian tubules lumen.

MT : Malpighian tubules.
MB : Muscle bundle.
PFB : Partial fat body.

(X = 400)

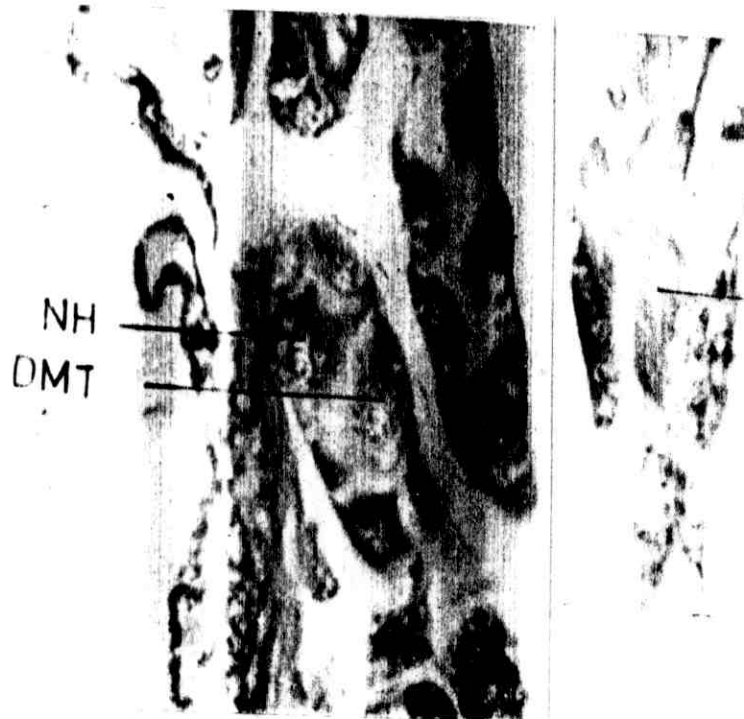


Fig. (17)



Fig. (18)

Fig.(19) : A transverse section of a larva treated with B. thuringiensis.

GL : Gut lumen.
RECM: Ruptured epithelium cell membrane.
NH : Nucleus hypertrophy.
TT : tracheal tube.
TM : tracheal matrix.
DMB : Destroyed muscle bundle.

Fig. (20): A longitudinal section in an infected larva treated with Bacillus sphaericus 1593 - 4.

DEC : Destroyed epithelial cell.
DMB : **Destroyed** muscle bundle.
DFT : Destroyed fat tissue.
GL : Gut lumen.

(X = 400)



Fig. (19)

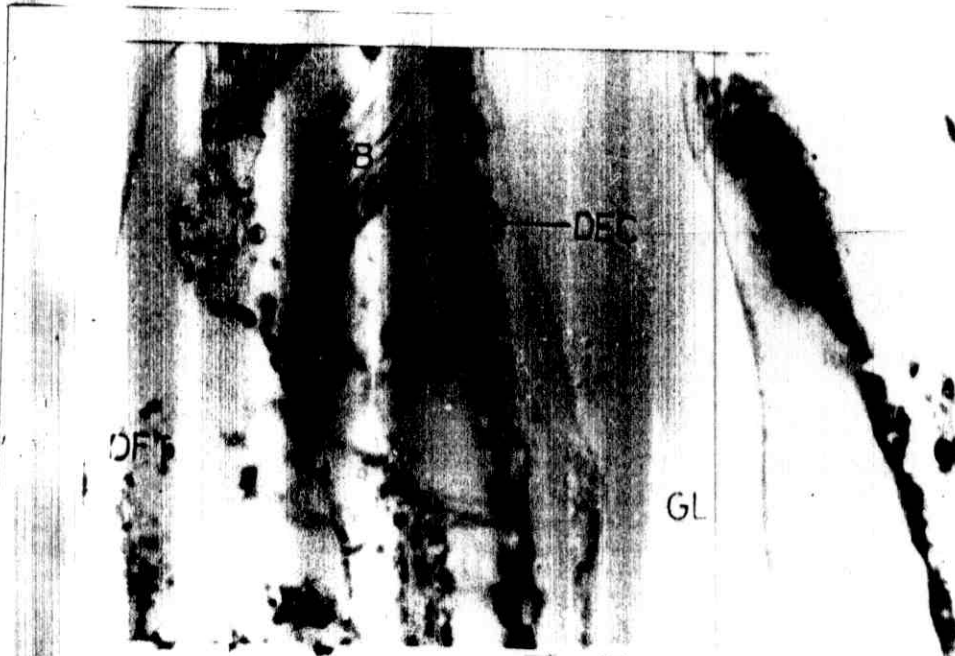


Fig. (20)

Fig. (21): A longitudinal section in an another infected larva treated with Bacillus sphaericus 1593-4.

DFT : **Destroyed** fat tissue.
GL : Gut lumen.
DEC : **Destroyed** epithelial cell.
VN : **Vacuolated** nucleus.
DMB : **Destroyed** muscle bundle.

(X = 400)

Fig. (22): A transverse section of a larva infected with Bacillus sphaericus. 1593-4.

TT : Tracheal tube.
DMB : **Destroyed** muscle bundle.
GL : Gut lumen.

(X = 400)



Fig. (21)

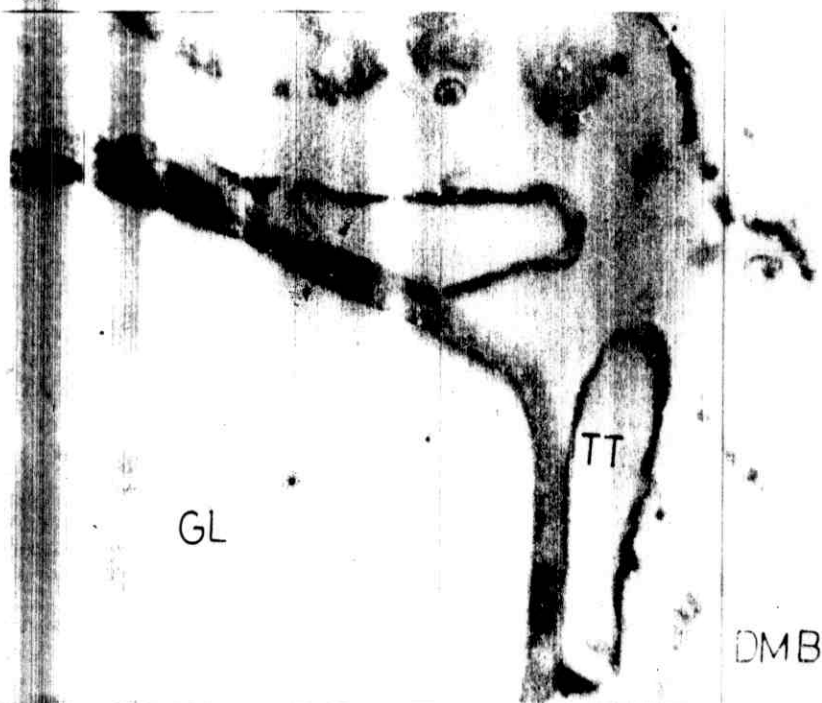


Fig. (22)

Fig. (23): A longitudinal section in an infected larva treated with the chemical larvicide dieldrin.

NH : Nucleus hypertrophy.
DPM : Destroyed peritrophic membrane.
GC : Gut Contents.
VC : **Vacuolated** cytoplasm.
DEC : Destroyed epithelial cell
DMB : Destroyed muscle bundle.

(X = 400)

Fig. (24): A transverse section in an infected larva treated with dieldrin through the thoracic region.

DCL : Destroyed cortical layer.
NM : Neuropile mass hypertrophy.
DMB : Destroyed muscle bundle.
NH : Nucleus hypertrophy.
EC : Epithelium cell.

(X = 400)

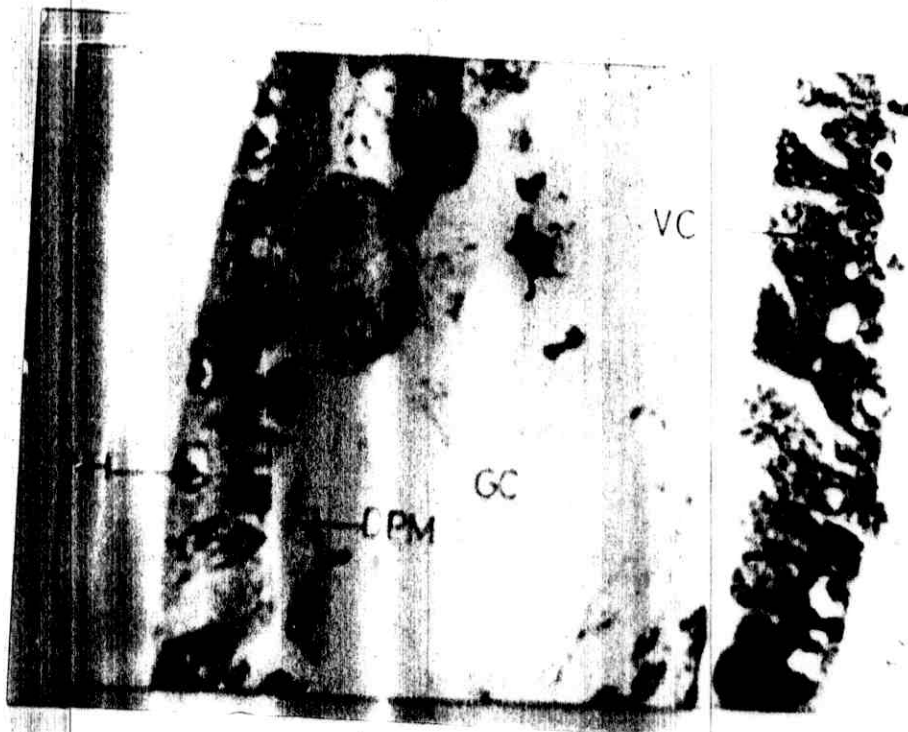


Fig. (23)



Fig. (24)

Fig. (25): A transverse section in a dieldrin treated larva.

GC : Gut contents.
DEC : Destroyed epithelial cell.
NH : Nucleus hypertrophy.
TT : Tracheal tube.
TM : Tracheal matrix.
DFT : Destroyed fat tissue.
DMB : Destroyed muscle bundle.

(X = 400)

Fig. (26): A longitudinal section of a dieldrin treated larva.

DMTC : Destroyed Cells of Malpighian tubules.

(X = 400).

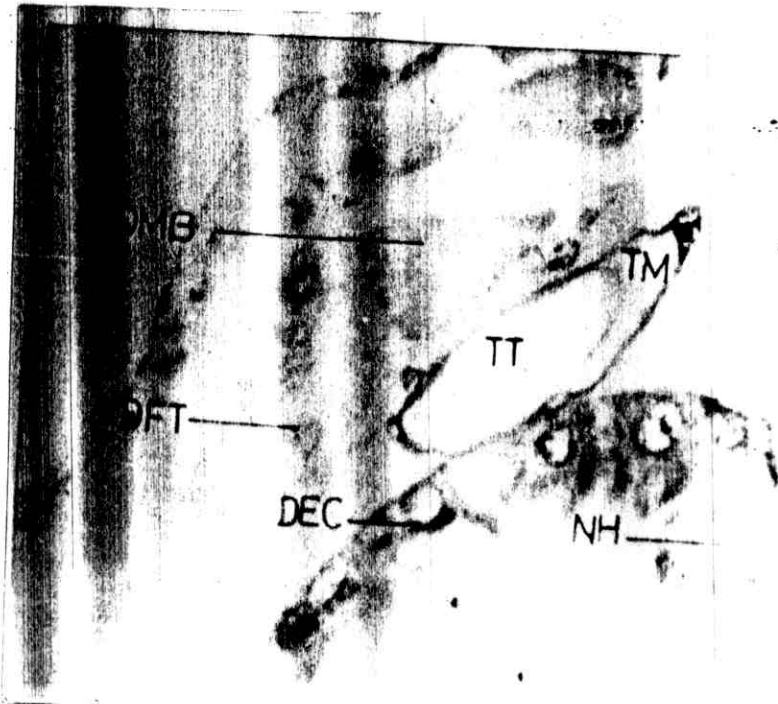


Fig. (25)



Fig. (26)

Fig. (27): A longitudinal section of a larva treated with (Bacillus thuringiensis H-14 + diadrin) mixture.

DMB : Destroyed muscle bundle.
ROC : Release of cytoplasm.
NH : Nucleus hypertrophy.
GC : Gut contents.
DEC : Destroyed epithelial cells.

(X = 400)

Fig. (28): A longitudinal section of a larva treated with (B. thuringiensis H-14 + dieldrin) mixture.

GC : Gut contents.
EC : Epithelial cells.

(X = 400)



Fig. (27)



Fig. (28)

Fig. (29): A longitudinal section in a larva treated with (Bacillus thuringiensis H-14 + dieldrin) mixture, showing the separation of the gut epithelium.

GC : Gut contents.

DCEC : Destroyed cytoplasm of epithelium cell.

SRC : Swollen regenerative cell.

(X = 400)

Fig. (30): A longitudinal section in a larva treated with (B. thuringiensis H-14 + dieldrin) mixture, **showing the vacuolated** apical parts of the gut epithelium.

GC : Gut contents.

DEC : Destroyed epithelial cell.

(X = 400)

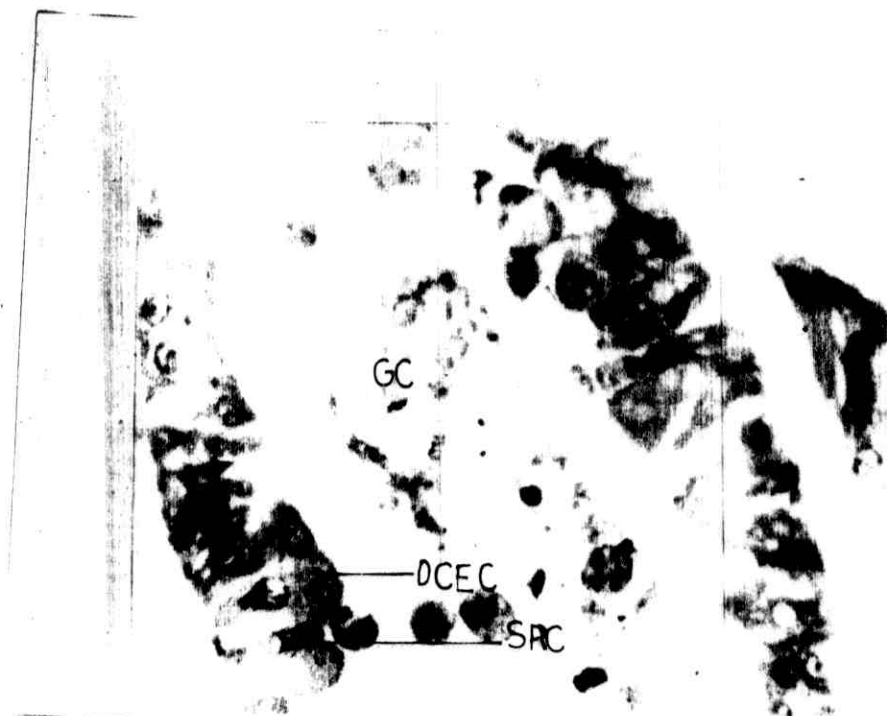


Fig. (29)

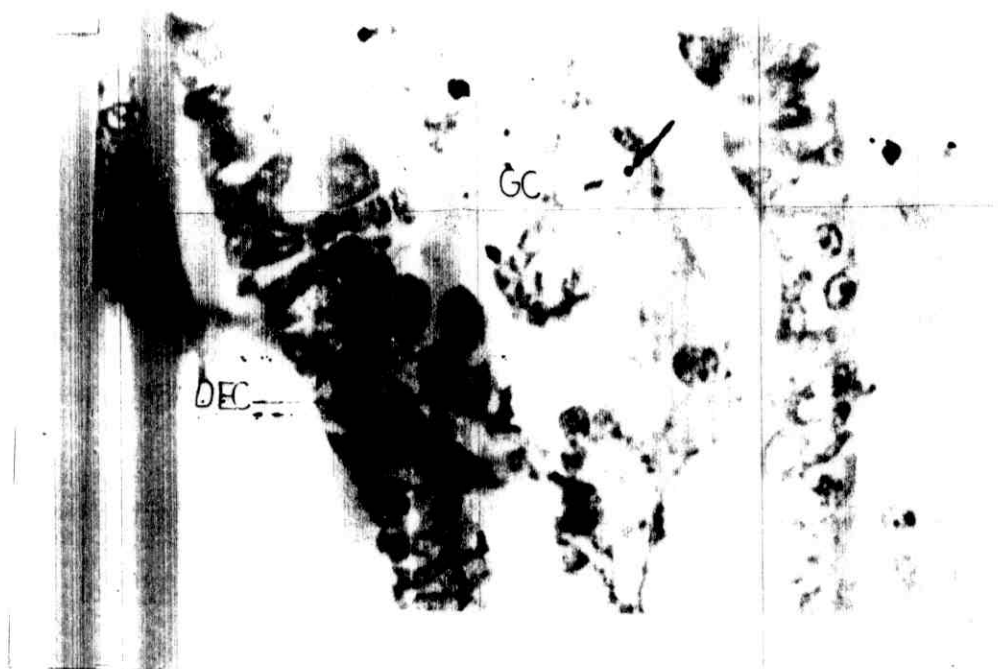


Fig. (30)

Fig. (31): A transverse section of a larva in early sign of infection treated with (Bacillus thuringiensis H-14 + dieldrin) mixture.

GL : Gut lumen.
EC : Epithelial cell.
(X = 400)

Fig. (32): A transverse section of a larva in advanced stage of infection, treated with (Bacillus thuringiensis H-14 + dieldrin) mixture.

N : Nucleus
GCa : Gastric caeca.
GC : Gut contents.
ROC : Release of cytoplasm.
EB : Empty apical portion.
NH : Nucleus hypertrophy.
DMB : Destroyed muscle bundle.
DFT : Destroyed fat tissue.

(X = 400)



Fig. (31)



Fig. (32)

Fig. (33): A longitudinal section with much higher magnification of a larva treated with (B. thuringiensis H-14 + dieldrin) mixture, showing the Ballo **on-shaped epithelial cell**.

N : Nucleus
BS : **Balloon** -Shaped epithelium cell.
EB : Empty apical portion.

(X = 600)

Fig. (34): A longitudinal section in a larva treated with (Bacillus sphaericus 1593-4)+ dieldrin) mixture.

AEC : Affected epithelium cell.
GC : Gut contents.
DMB : Destroyed muscle bundle.

(X = 400)

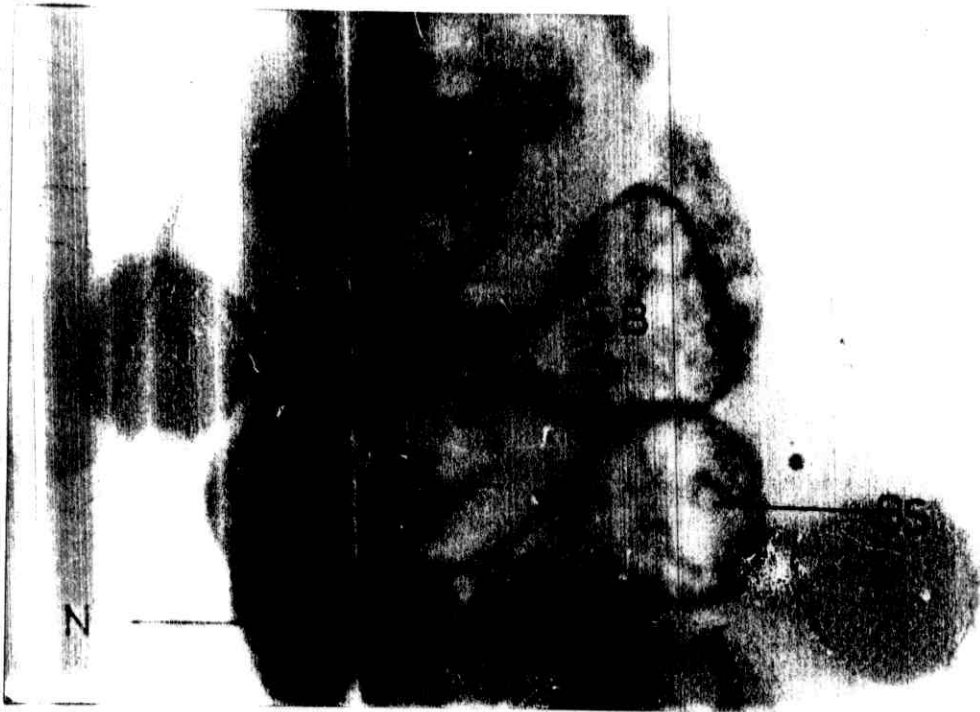


Fig. (33)



Fig. (34)

Fig. (35): A longitudinal section in a larva treated with (Bacillus sphaericus & dieldrin) mixture.

CH : Cell hypertrophy.
SEC : Swallen epithelial cell.
DPM : Destroyed peritrophic membrane.
GC : Gut contents.
NH : Nucleus hypertrophy.

(X = 400)

Fig. (36): A longitudinal section in a larva treated with (Bacillus sphaericus & Dieldrin) mixture. Posterior part of mid-gut region.

N : Nucleus hypertrophy.
GC : Gut contents.
RCM : Rupture cell membrane.
ROC : Release of cytoplasm.

(X = 400)

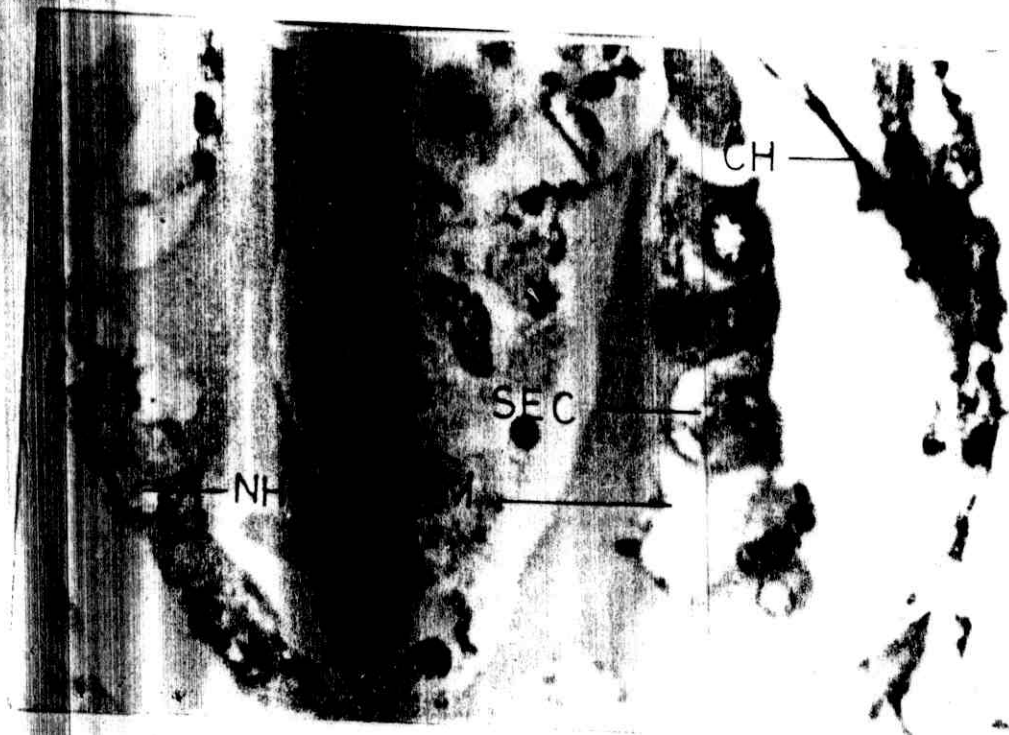


Fig. (35)

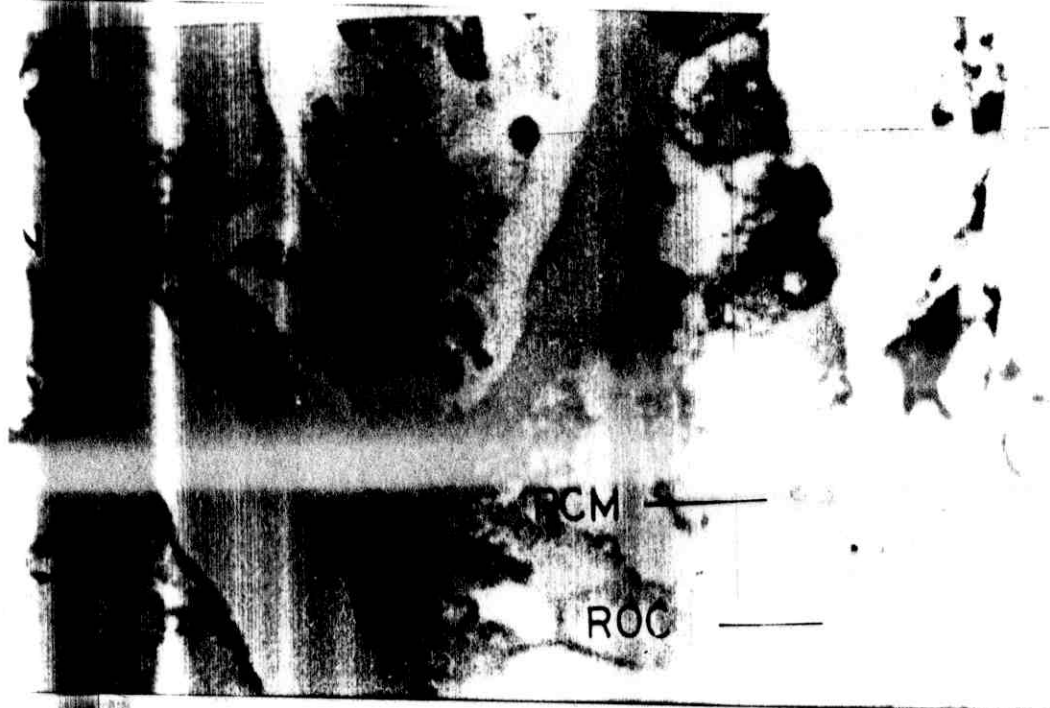


Fig. (36)

Fig. (37): A transverse section of a larva treated with (Bacillus sphaericus & dieldrin) mixture.

GC : Gut contents.
DEC : Destroyed epithelial cell.
DMB : Destroyed muscle bundle.

(X = 200)

Fig. (38): A transverse section of a larva treated with (Bacillus sphaericus & dieldrin) mixture.

VN : **Vacu**olated nucleus.
GL : Gut lumen.
SEC : Separate spindle shaped epithelium cell.

(X = 400)

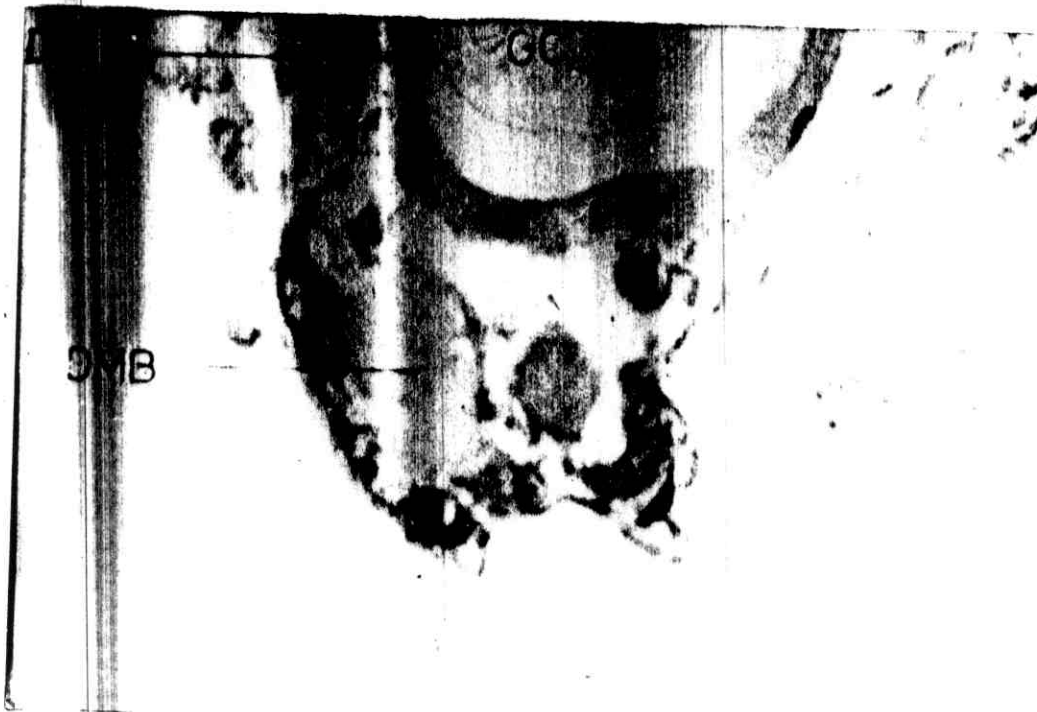


Fig. (37)



Fig. (38)

Fig. (39): A transverse section of a larva treated with (Bacillus sphaericus & dieldrin) mixture.

VC : Vacuolated cytoplasm.
NH : Nucleus hypertrophy.
G C : Gut contents.
DPM : Destroyed peritrophic membrane.
TT : Tracheal tube.
TM : Tracheal matrix.

(X = 400)

Fig. (40): A transverse section of a larva treated with (Bacillus sphaericus & dieldrin) mixture. Post-mortum (just died).

GCa : Gastric caeca.
N : Nucleus hypertrophy.
GC : Gut contents.
EC : Epithelial cell.
TT : Tracheal tube.
TM : Tracheal matrix.

(X = 400)

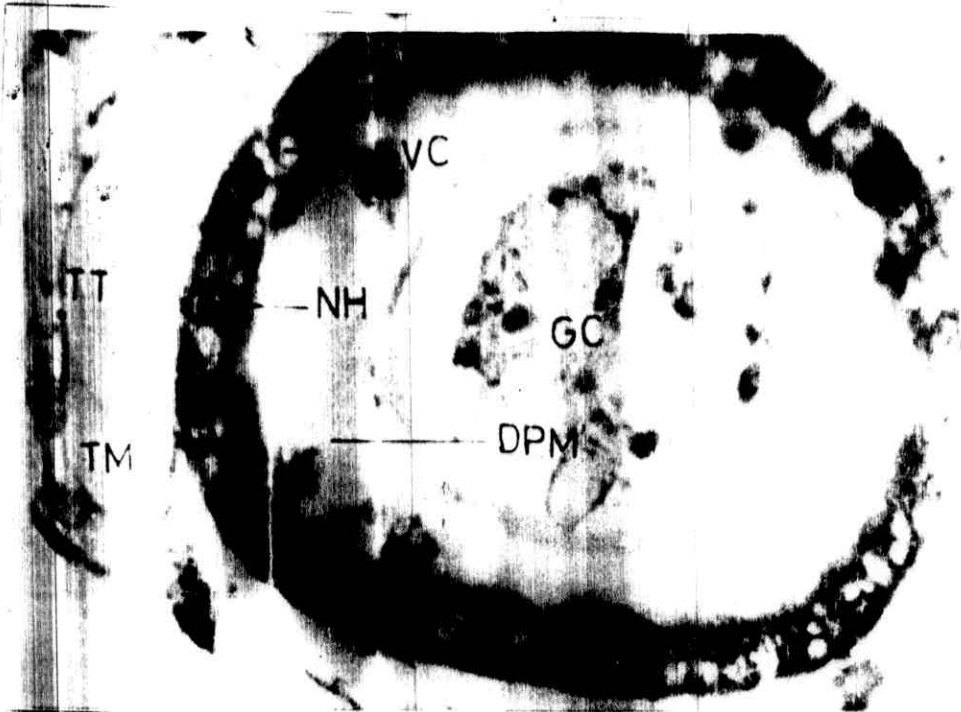


Fig. (39)

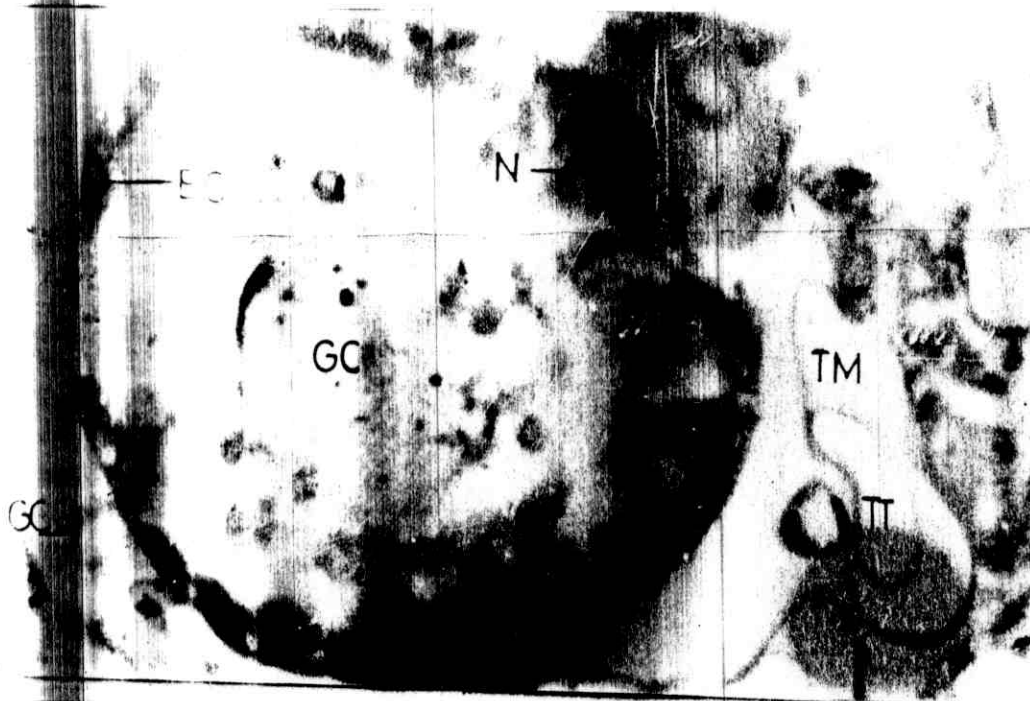


Fig. (40)

Fig. (41): A longitudinal section of a larva treated with (Bacillus thuringiensis H-14 & B. sphaericus) mixture.

DMB : Destroyed muscle bundle .
NH : Nucleus hypertrophy.
GC : Gut contents.
AEC : Affected epithelium cell.

(X = 400)

Fig. (42): A longitudinal section of a larva treated with (Bacillus thuringiensis H-14 & B. sphaericus) mixture. Showing the epithelial gut celles with transparent apical parts.

FT : Fat tissue.
SEC : Swallen epithelium cell.
GL : Gut lumen.
MB : Muscle bundle.

(X = 400)



Fig. (41)



Fig. (42)

Fig. (43): A longitudinal section of a larva infected with (Bacillus thuringiensis H-14 & B. sphaericus) mixture. Showing the ballo on-shaped cell.

BS : Balloon-shaped epithelial cell.
MB : Muscle bundle.

(X = 400)

Fig. (44): A longitudinal section of a larva infected with (Bacillus thuringiensis H-14 & B. sphaericus) mixture. Advanced stage in infection.

M : Muscle bundle.
PM : Peritrophic membrane.
GC : Gut contents.
DEC : Destroyed epithelium cell.

(X = 400)



Fig. (43)

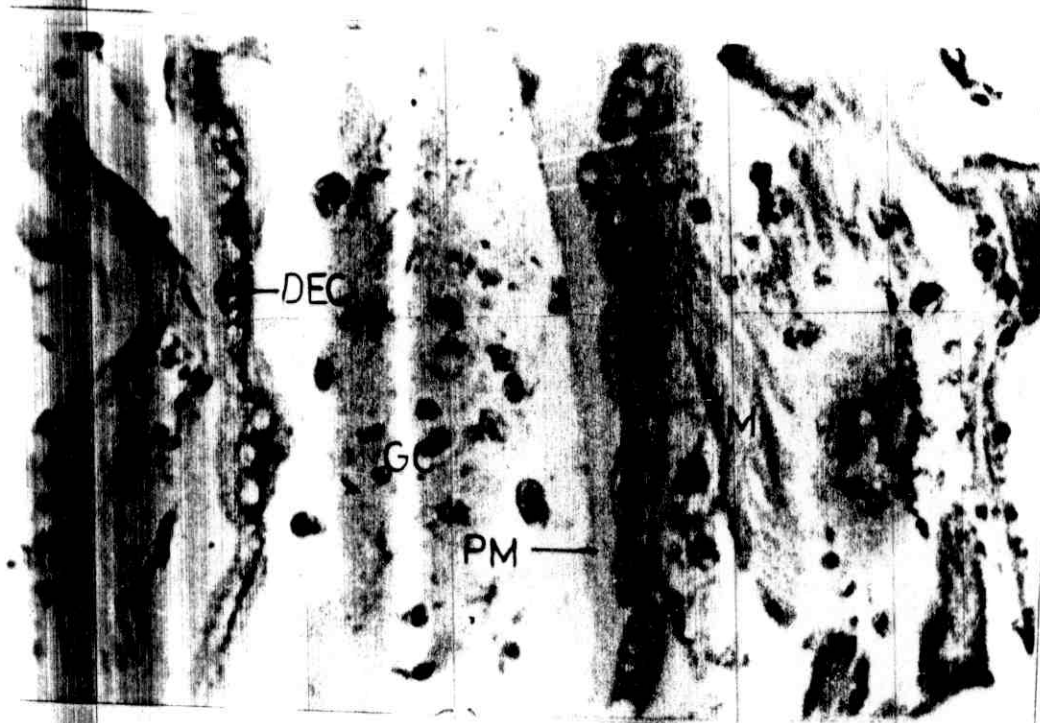


Fig. (44)

Fig. (45): A longitudinal section in an infected larva (post-mortium, just died) with (Bacillus thuringiensis H-14 & Bacillus sphaericus) mixture.

VC : Vacuolated cytoplasm.
NH : Nucleus hypertrophy.
GL : Gut lumen.
DM : Destroyed Muscle bundle.
(X = 400)

Fig. (46): A longitudinal section in a larva treated with (Bacillus thuringiensis H-14 & B. sphaericus) mixture.

SEC : Separate epithelium cell.
GC : Gut contents.

(X = 400).

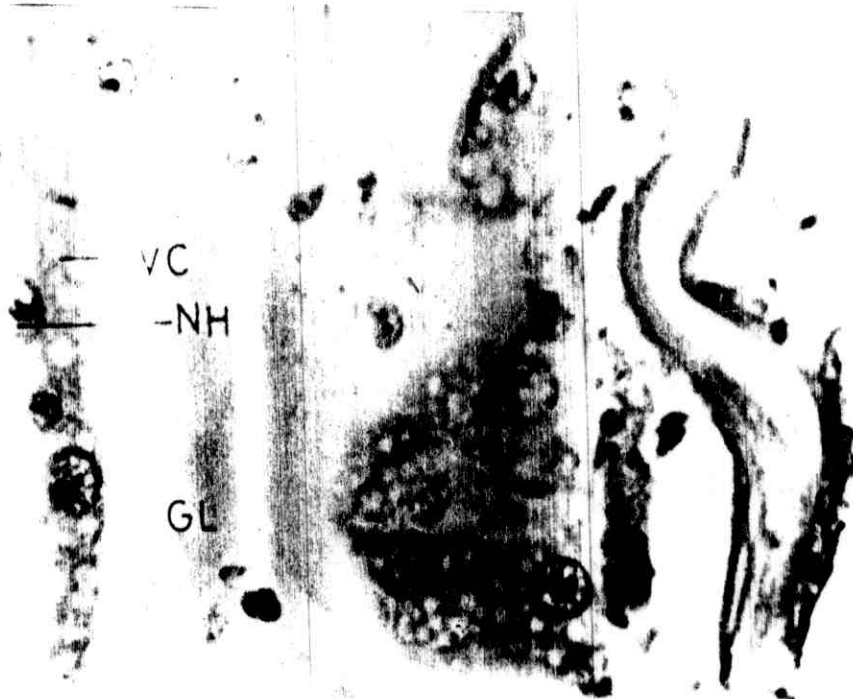


Fig. (45)

