### **RESULTS AND DISCUSSION**

# Laboratory evaluation of three B.thuringiensis products against different larval instar of S. littoralis:

#### 1.1. Effect on larval mortality:

#### 1.1.a. Newly hatched:

From data in table (1), it is clear that the mortality percentage after 5 days of larval feeding on treated castor-bean leaves was a concentration dependent, *i.e.*, mortality % increased by raising the applied concentration of bioinsecticide.

The corrected mortality percentages after Dipel 2X treatment ranged from 26.7 % by applying the lowest concentration  $(0.2 \times 10^3 \text{ IU} / \text{mg})$  to 100.0 % by using the highest concentration  $(12.8 \times 10^3 \text{ IU/ mg})$ .

By using the same concentrations of Dipel EC , the obtained moralities ranged also between 26.7 and 100.0 %, respectively. While, in case of Ecotech, the applied concentrations ranged from  $0.22 \times 10^3$  to  $14.4 \times 10^3$  IU/mg and subcquently, the corrected mortality percentages recorded also 26.7 - 100.0 % , respectively (Table , 1). The concentration – mortality lines are graphically illustrated in Fig.(1) and confirmed the same results concerning the obtained mortality percentages. Form data of this figure and those tabulated in Table (2), the LC50's after 5 days of treatment were  $0.464 \times 10^3$  IU/mg (with confidence limites of  $0.298 \times 10^3$  and  $0.649 \times 10^3$  IU/mg at 95% probability) for Dipel 2X ,  $0.889 \times 10^3$  IU/mg (confidence limits  $0.5920 \times 10^3$  and  $1.257 \times 10^3$  IU /mg ) for Dipel EC and  $0.810 \times 10^3$  IU/mg  $(0.538 \times 10^3 - 1.134 \times 10^3$  IU/mg) for Ecotech treatment .

Table (1): Corrected mortality percentages among the different larval instar of S. littoralis, 5 days after treatment with three commercial bioinsecticides.

3	Newly hatched	ched	Ist		2 <sup>nd</sup>		3 <sup>rd</sup>	1	4 <sup>th</sup>		5 <sup>th</sup>		6 <sup>th</sup>	
теаннен	Con.	Mort.	Con.	Mort.	Con.	Mort.	Con.	Mort.	Con.	Mort.	Con.	Mort.	Con.	Mort.
Dipel 2X	12.8x10 <sup>3</sup>	100.0	$12.8 \times 10^{3}$	96.7	19.2 x 10 <sup>3</sup>	100.0	$24 \times 10^{3}$	96.7	$32.0 \times 10^3$	96.7	$40.0 \times 10^{3}$	93.3	$48.0 \times 10^{3}$	90.0
	$6.4 \times 10^3$	96.7	$6.4 \times 10^3$	83.3	$9.6 \times 10^{3}$	93.3	$12 \times 10^{3}$	83.3	$16.0 \times 10^3$	93.3	$20.0 \times 10^{3}$	86.7	$24.0 \times 10^{3}$	80.0
	$3.2 \times 10^3$	86.7	$3.2 \times 10^{3}$	76.7	$4.8 \times 10^{3}$	86.7	$6 \times 10^{3}$	76.7	$8.0 \times 10^{3}$	86.7	$10.0 \times 10^3$	70.0	$12.0 \times 10^3$	66.7
	$1.6 \times 10^3$	73.3	$1.6 \times 10^{3}$	53.3	$2.4 \times 10^3$	60.0	$3 \times 10^{3}$	63.3	$4.0 \times 10^{3}$	73.3	$5.0 \times 10^{3}$	56.7	$6.0 \times 10^3$	53.3
	$0.8 \times 10^{3}$	60.0	$0.8 \times 10^{3}$	40.0	$1.2 \times 10^{3}$	53.3	$1.5 \times 10^{3}$	53.3	$2.0 \times 10^{3}$	53.3	$2.5 \times 10^3$	46.7	$3.0 \times 10^{3}$	36.7
	$0.4 \times 10^3$	56.7	$0.4 \times 10^{3}$	23.3	$0.6 \times 10^3$	40.0	$0.75 \times 10^3$	36.7	$1.0 \times 10^{3}$	36.7	$1.25 \times 10^3$	23.3	$1.5 \times 10^{3}$	26.7
	$0.2 \times 10^3$	26.7			$0.3 \times 10^{3}$	20.0	$0.37x\ 10^3$	26.7	$0.5 \times 10^3$	26.7	$0.62 \times 10^3$	13.3	$0.75 \times 10^3$	10.0
Dipel EC	$12.8 \times 10^3$	100.0	$12.8 \times 10^3$	96.7	$19.2 \times 10^3$	100.0	25.6x10 <sup>3</sup>	96.7	$35.2 \times 10^3$	96.7	$41.6 \times 10^3$	90.0	$56.0 \times 10^3$	86.7
	$6.4 \times 10^3$	83.3	$6.4 \times 10^3$	80.0	$9.6 \times 10^3$	93.3	12.8x 10 <sup>3</sup>	83.3	$17.6 \times 10^3$	90.0	$20.8x\ 10^3$	80.0	$28 \times 10^{3}$	73.3
Zwenie sa	$3.2 \times 10^{3}$	73.3	$3.2 \times 10^{3}$	76.7	$4.8 \times 10^{3}$	66.7	$6.4 \times 10^3$	73.3	$8.8 \times 10^{3}$	83.3	$10.4 \times 10^3$	73.3	$14.0x\ 10^3$	63.35
	$1.6 \times 10^3$	56.7	$1.6 \times 10^3$	56.7	$2.4 \times 10^3$	53.3	$3.2 \times 10^3$	50.0	$4.4 \times 10^3$	76.7	$5.2 \times 10^3$	53.3	$7.0 \times 10^3$	53.3
	$0.8 \times 10^{3}$	40.0	$0.8 \times 10^{3}$	36.7	$1.2 \times 10^3$	40.0	$1.6 \times 10^3$	46.7	$2.2 \times 10^{3}$	50.0	$2.6 \times 10^3$	40.0	$3.5 \times 10^3$	33.3
	$0.4 \times 10^3$	36.7	$0.4 \times 10^3$	30.0	$0.6 \times 10^3$	33.3	$0.8 \times 10^3$	36.7	$1.1 \times 10^{3}$	26.7	$1.3 \times 10^{3}$	20.0	$1.75 \times 10^3$	23.3
	$0.2 \times 10^3$	26.7	$0.2 \times 10^3$	26.7	$0.3 \times 10^3$	26.7	$0.4 \times 10^3$	20.0	$0.55 \times 10^3$	13.3	$0.65 \times 10^3$	13.3	$0.87 \times 10^3$	6.7
Ecotech	$14.4 \times 10^3$	100.0	$14.4 \times 10^3$	93.3	$21.6 \times 10^3$	93.3	$27.0 \times 10^3$	90.0	$36.0 \times 10^3$	100.0	$45.0 \times 10^3$	93.3	$63.0 \times 10^3$	90.0
	$7.2 \times 10^3$	93.3	$7.2 \times 10^3$	83.3	10.8x 10 <sup>3</sup>	86.7	$13.5 \times 10^3$	86.0	18.0x 10 <sup>3</sup>	93.3	$22.5 \times 10^3$	83.3	$31.5 \times 10^3$	80.0
	$3.6 \times 10^3$	76.7	$3.6 \times 10^3$	70.0	$5.4 \times 10^3$	73.3	$6.7 \times 10^3$	73.3	$9.0 \times 10^{3}$	66.7	11.25x10 <sup>3</sup>	70.0	15.75x10 <sup>3</sup>	63.3
	$1.8 \times 10^{3}$	53.3	$1.8 \times 10^{3}$	53.3	$2.7 \times 10^3$	60.0	$3.37 \times 10^3$	66.7	$4.5 \times 10^{3}$	53.3	$5.62 \times 10^3$	53.7	$7.87 \times 10^3$	56.7
	$0.9x\ 10^3$	50.0	$0.9 \times 10^3$	40.0	$1.35 \times 10^{3}$	53.3	1.68x 10 <sup>3</sup>	43.3	$2.25 \times 10^{3}$	40.0	$2.81 \times 10^{3}$	46.7	$3.93 \times 10^3$	36.7
	$0.45 \times 10^3$	43.3	$0.45 \times 10^3$	36.7	$0.67 \times 10^3$	33.3	$0.84 \times 10^3$	36.7	$1.12 \times 10^3$	20.0	$1.40 \text{x} 10^3$	20.0	$1.96 \times 10^3$	30.0
	$0.22 \times 10^3$	26.7	$0.22 \times 10^3$	20.0	$0.33 \times 10^3$	26.7	$0.42 \times 10^3$	20.0	$0.56 \times 10^3$	13.3	$0.70 \times 10^3$	16.7	$0.98 \times 10^3$	10.0

Table (2): LC<sub>50</sub> and LC<sub>90</sub> of B. thuringiensis in three commercial products experimented on different larval instars of S. littoralis after 5 days feeding on treated castor bean leaves.

				95% confi	95% confidence limets		95% confi	95% confidence limets
Larvarinstar	Пейшен	Siope	LC50	Lower	Upper	1000	Lower	Upper
Newly hatched	Dipel 2 X	1.47 ± 0.21	0.464x 10 <sup>3</sup>	$0.298 \times 10^3$	$0.649 \times 10^3$	$3.453 \times 10^3$	$2.282 \times 10^{3}$	6.544 x 10 <sup>3</sup>
	Dipel EC	$1.26 \pm 0.18$	$0.899 \times 10^3$	$0.592 \times 10^3$	$1.258 \times 10^3$	$9.258 \times 10^3$	$5.470 \times 10^3$	$21.582 \times 10^3$
	Ecotech	$1.34 \pm 0.18$	$0.810 \times 10^{3}$	$0.538 \times 10^3$	$1.134 \times 10^3$	$7.358 \times 10^3$	4.570 x 10 <sup>3</sup>	15.559 x 10 <sup>3</sup>
1 <sup>st</sup> instar	Dipel 2X	$1.24 \pm 0.17$	$0.998 \times 10^{3}$	0.670 x10 <sup>3</sup>	$1.416 \times 10^3$	$8.125 \times 10^3$	$5.232 \times 10^3$	$16.688 \times 10^3$
	Dipel EC	$1.56 \pm 0.23$	$1.226 \times 10^3$	$0.842 \times 10^3$	$1.667 \times 10^3$	$10.789 \times 10^3$	$6.245 \times 10^3$	$26.158 \times 10^3$
	Ecotech	$1.2 \pm 0.17$	$1.188 \times 10^{3}$	$0.792 \times 10^3$	1.699 x 10 <sup>3</sup>	13.862 x 10 <sup>3</sup>	$7.805 \times 10^3$	$35.627 \times 10^3$
2 <sup>nd</sup> instar	Dipel 2X	$1.57 \pm 0.20$	$1.06 \times 10^{3}$	$0.746 \times 10^3$	$1.427 \times 10^3$	$6.931 \times 10^3$	$4.627 \times 10^3$	$12.714 \times 10^3$
	Dipel EC	$1.41 \pm 0.18$	$1.528 \times 10^3$	$1.069 \times 10^3$	$2.882 \times 10^{3}$	$12.296 \times 10^3$	$7.686 \times 10^3$	25.3872 x10 <sup>3</sup>
	Ecotech	$1.16 \pm 1.17$	$1.352 \times 10^3$	$0.855 \times 10^3$	1.960 x 10 <sup>3</sup>	$17.037 \times 10^3$	$9.634 \times 10^3$	43.981 x 10 <sup>3</sup>
3 <sup>rd</sup> instar	Dipel 2X	$1.22 \pm 0.17$	$1.622 \times 10^3$	$1.053 \times 10^3$	$2.317 \times 10^{3}$	$18.019 \times 10^{3}$	$10.541 \times 10^3$	$43.098 \times 10^3$
	Dipel Ec	$1.29 \pm 0.18$	$2.011 \times 10^3$	1.384 x10 <sup>3</sup>	$2.816 \times 10^{3}$	$19.520 \times 10^3$	$11.576 \times 10^3$	$44.813 \times 10^3$
	Ecotec	$1.24 \pm 0.17$	$2.077 \times 10^{3}$	$1.393 \times 10^3$	$2.945 \times 10^3$	22.320 x 10 <sup>3</sup>	12.958 x 10 <sup>3</sup>	53.791 x10 <sup>3</sup>
4 <sup>th</sup> instar	Dipel 2X	$1.49 \pm 0.19$	$1.843 \times 10^{3}$	$1.264 \times 10^3$	$2.515 \times 10^{3}$	$13.369 \times 10^3$	$8.758 \times 10^{3}$	$25.475 \times 10^3$
	Dipel EC	$1.72 \pm 0.21$	$2.267 \times 10^3$	$1.678 \times 10^3$	$2.982 \times 10^{3}$	$12.568 \times 10^3$	$8.603 \times 10^3$	$21.917 \times 10^3$
	Ecotech	$1.80 \pm 0.21$	$3.654 \times 10^3$	$2.779 \times 10^3$	$4.766 \times 10^3$	18.826 x 10 <sup>3</sup>	12.924 x 10 <sup>3</sup>	$32.501 \times 10^3$
5 <sup>th</sup> instar	Dipel 2X	$1.49 \pm 0.18$	$4.144 \times 10^3$	$3.014 \times 10^3$	$5.616 \times 10^3$	$30.045 \times 10^3$	$19.024 \times 10^3$	$60.173 \times 10^3$
	Dipel EC	$1.43 \pm 0.18$	$4.691 \times 10^3$	$3.400 \times 10^3$	$6.445 \times 10^3$	$37.059 \times 10^3$	$22.638 \times 10^3$	$79.369 \times 10^3$
	Ecotec	$1.42 \pm 0.18$	$4.694 \times 10^3$	$3.379 \times 10^3$	6.437 x 10 <sup>3</sup>	$37.379 \times 10^3$	$23.002 \times 10^3$	$79.070 \times 10^3$
6 <sup>th</sup> instar	Dipel 2X	$1.39 \pm 0.18$	$5.939 \times 10^3$	$4.288 \times 10^3$	$8.218 \times 10^{3}$	$48.976 \times 10^3$	$29.408 \times 10^3$	$108.352 \times 10^3$
	Dipel EC	$1.4 \pm 0.18$	$8.477 \times 10^3$	$6.133 \times 10^3$	$11.878 \times 10^{3}$	$72.387 \times 10^3$	$42.147 \times 10^3$	$170.035 \times 10^3$
	Ecotech	$1.35 \pm 0.18$	$7.574 \times 10^3$	5.409 x 10 <sup>3</sup>	$10.575 \times 10^3$	$67.671 \times 10^3$	39.785 x10 <sup>3</sup>	156.506 x10 <sup>3</sup>

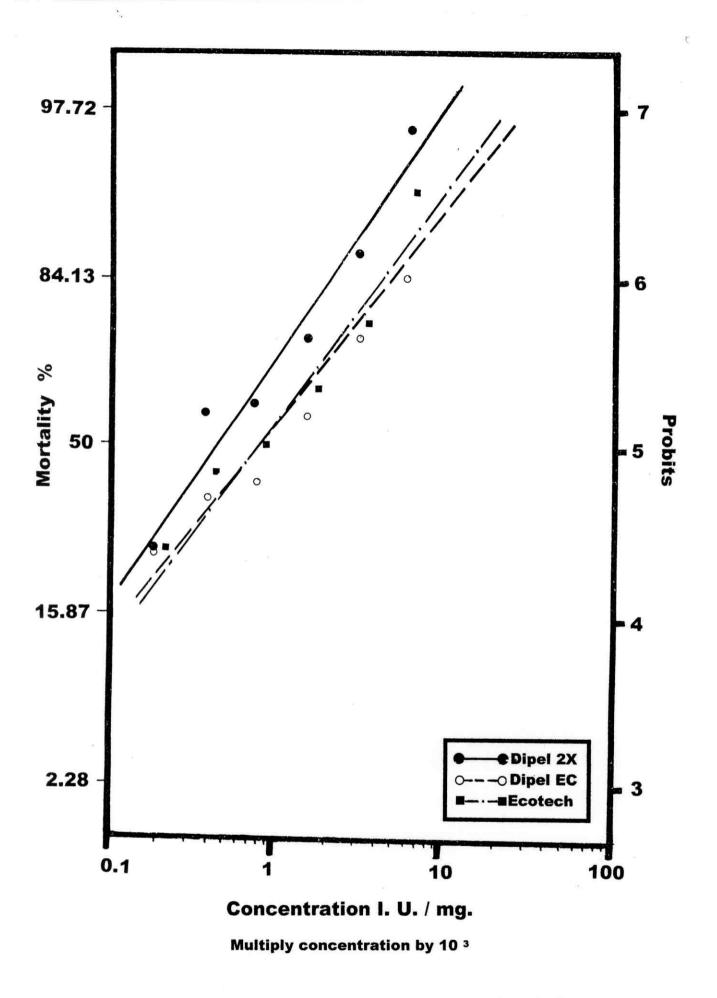


Fig. (1): Regression lines indicating mortalities of *S. littoralis* larvae, after 5 days of feeding the newly hatched larvae on castor – bean leaves treated with three bioinsecticides.

....

# 1.1.b. Against 24 h. old 1st instar Larvae :

As shown in Table (1), the mortality percentages ranged from 23.3 - 96.7, 26.7 - 96.7 and 20.0 - 93.3%, when the *S.littoralis* first instar larvae were fed on castor-bean leaves treated with different concentration of Dipel 2X, Dipel EC and Ecotech , respectively (Table,1). While, data illustrated in (Fig.2) and those presented in Table (2) indicate that the respective values of LC<sub>50</sub> after 5 days of treatment by the mentioned bioinsecticides were  $0.998 \times 10^3$  IU/mg (confidence limits;  $0.670 \times 10^3 - 1.416 \times 10^3$  IU /mg),  $1.226 \times 10^3$  ( $0.842 \times 10^3 - 1.667 \times 10^3$ ) IU/ mg and  $1.188 \times 10^3$  ( $0.792 \times 10^3 - 1.699 \times 10^3$ ) IU/ mg at 95% probability, respectively.

# 1.1.c. Against 2<sup>nd</sup> instar larvae :

After 2 days feeding of *S.littoralis*  $2^{nd}$  instar larvae on castor – bean leaves treated with different concentrations of bioinsectcide followed by 3 days feeding on fresh untreated castor bean leaves, the corrected mortality percentages were 20.0, 26.7 and 26.7% by applying the bioinsecticides at their lowest concentrations  $(0.3 \times 10^3 \, , 0.3 \times 10^3 \, \text{and} \, 0.33 \times 10^3 \, \text{IU/mg}$  of Dipel 2X , Dipel EC and Ecotech, respectively; Table,1). While, treating of leaf discs by the mentioned compounds their highest concentrations  $(19.2 \times 10^3 \, , 19.2 \times 10^3 \, \text{and} \, 21.6 \times 10^3 \, \text{IU/mg}$ ) caused 100, 100 and 93.3 % mortalities, respectively (Table, 1).

At 95 % probability level, the respective  $LC_{50}$ 's of the mentioned compounds after 5 days of treatment were  $1.06x10^3$  ( $0.746x10^3$ - $1.427x10^3$ ) IU /mg in case of Dipel 2X ,  $1.528x10^3$  ( $1.069x10^3$  –  $2.882x10^3$ ) IU/mg for Dipel Ec and  $1.352 \times 10^3$  IU/ mg. ( $0.855 \times 10^3$  –  $1.960 \times 10^3$ ) IU/mg in case of Ecotech treatment (Table, 2 and Fig.3) .

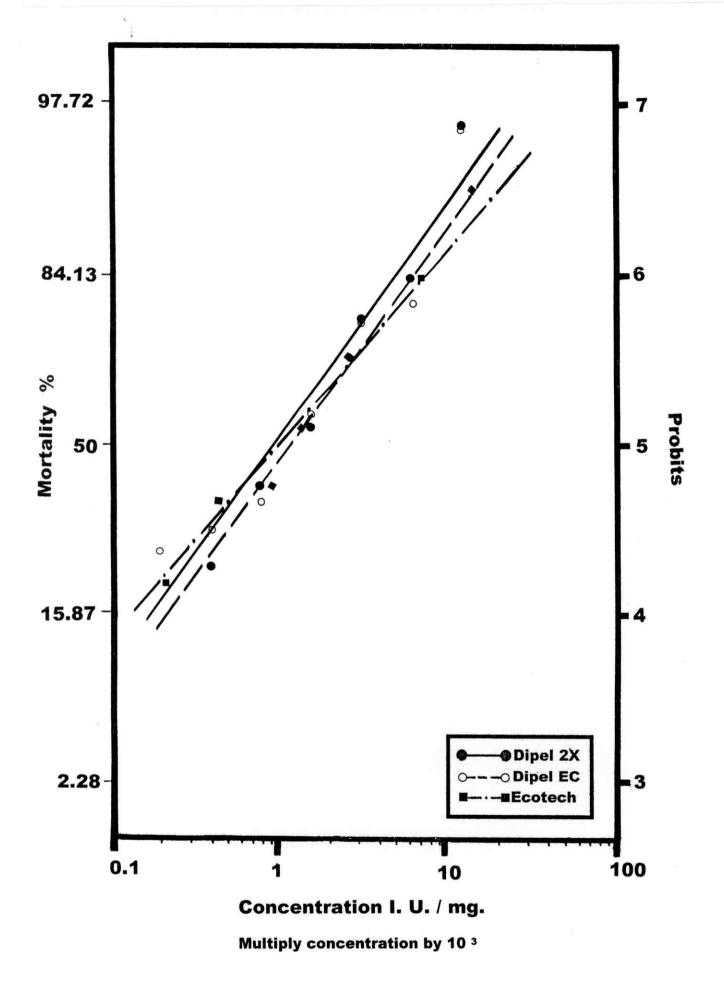


Fig. (2): Regression lines showing mortality percentages of S.

littoralis among the larvae fed in their 1<sup>st</sup> instar on castorbean leaves treated with three bioinsecticides.

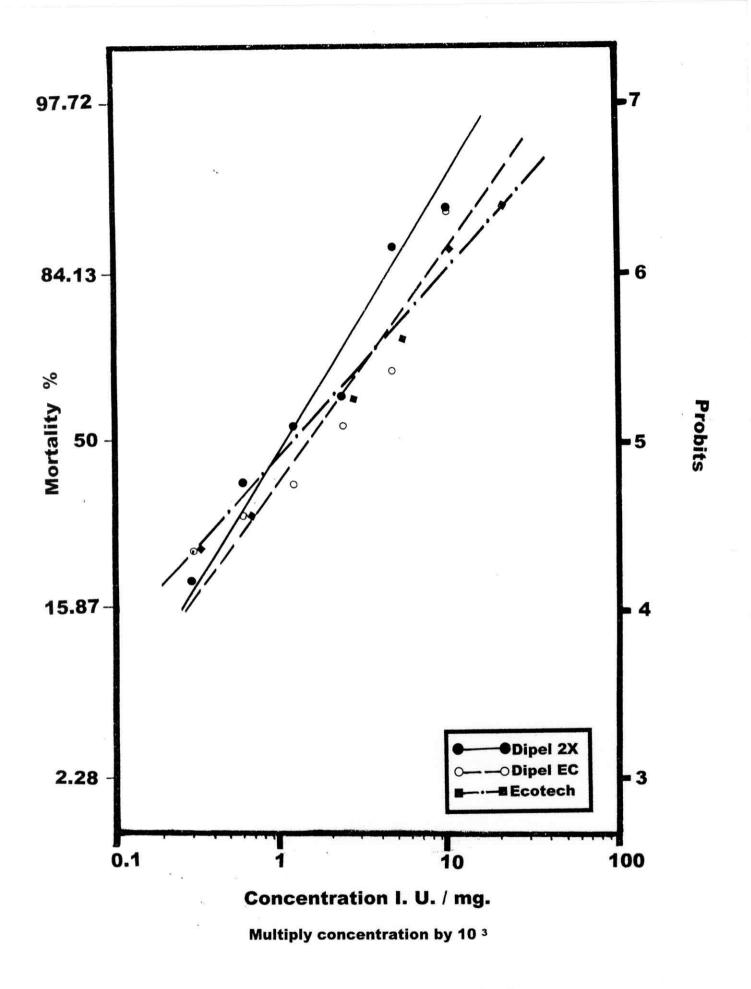


Fig. (3): Regression lines showing mortalities of *S. littoralis* due to feeding the 2<sup>nd</sup> larvae instar on castor – bean leaves treated with three bioinsecticides.

# 1.1.d. Against 3rd larval instar of S. littoralis:

From data in Table (1), it is clear that the recorded corrected mortality percentages ranged from 26.7- 96.7 % (by feeding on leaf – discs treated with Dipel 2X at concentrations  $0.37x10^3 - 24 \times 10^3$  IU/mg, respectively), 20.0 - 96.7 %  $(0.4x10^3 - 25.6x10^3$  IU/mg of Dipel EC) and 20.0-90.0 % (for concentrations of  $0.42x10^3$ - $27.0x10^3$  IU/mg of Ecotech) by applying the bioinsecticides at their lowest and highest concentrations, respectively. The LC<sub>50</sub>'s after 5 days of treatment with Dipel 2X, Dipel EC and Ecotech were  $1.622x10^3$  ( $1.053x10^3$ - $2.317x10^3$ ) IU/mg,  $2.011 \times 10^3$  ( $1.384x10^3$ - $2.816x10^3$  IU/mg and  $2.077x \times 10^3$  IU/mg. ( $1.393x10^3$ - $2.945x10^3$  IU/mg at 95% confidence limits), respectively (Table,2 and Fig.4).

#### 1.1.e. Fourth larval instar treatments:

After 5 days of *S.littoralis*  $4^{th}$  larval instar treatment by Dipel 2X, Dipel EC and Ecotech, the recorded corrected mortality percentages ranged from a minimum of 26.7% to maximum of 96.7% by feeding on the lowest  $(0.5 \times 10^3 \text{ IU/mg})$  and highest  $(32 \times 10^3 \text{ IU/mg})$  concentrations of Dipel 2X, respectively.

As for Dipel EC and Ecotech treatments, these percentages were 13.3- 96.70% ( $0.55\times10^3$  and  $35.2\times10^3$  IU/mg), and 13.3-100% ( $0.56\times10^3$  and  $36.0\times10^3$  IU/mg), respectively (Table, 1). Data in Table (2) and Fig. (5) show that after 5 days of Dipel 2X, Dipel EC and Ecotech treatments, the LC<sub>50</sub>'s were  $1.843\times10^3$  ( $1.264\times10^3-2.515\times10^3$  IU/mg.),  $2.267\times10^3$  ( $1.678\times10^3-2.982\times10^3$  IU/mg) and  $3.654\times10^3$  ( $2.779\times10^3-4.766\times10^3$  IU/mg. at 95% confidence limits), respectively.

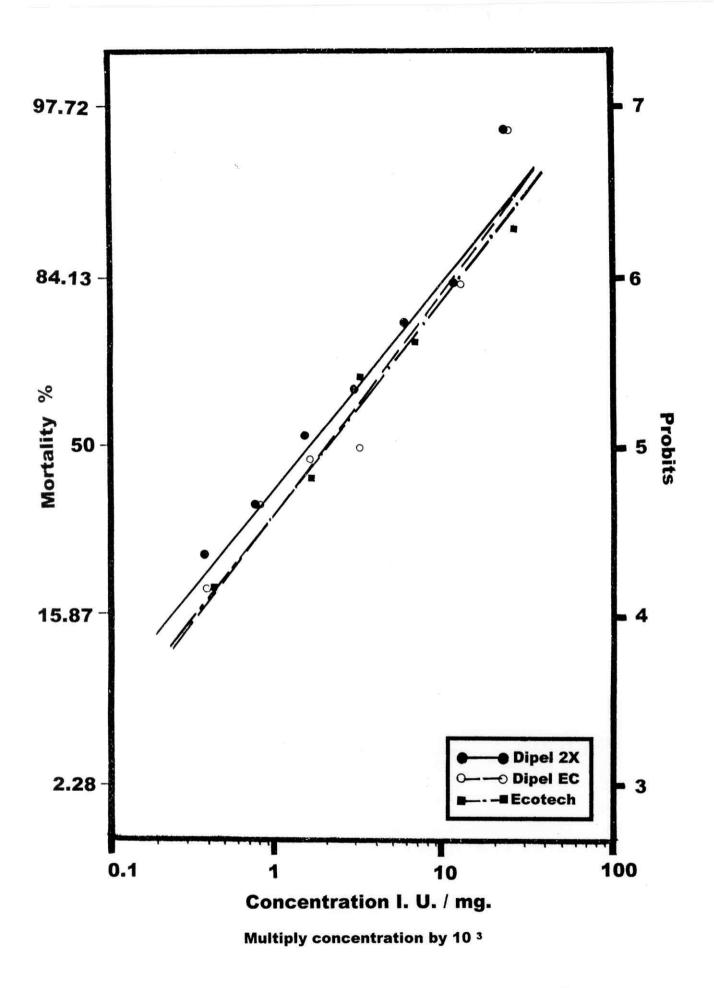


Fig. (4): Regression mortality lines after of *S. littoralis* 3<sup>rd</sup> instar larval feeding on castor – bean leaves treated with three bioinsecticides.

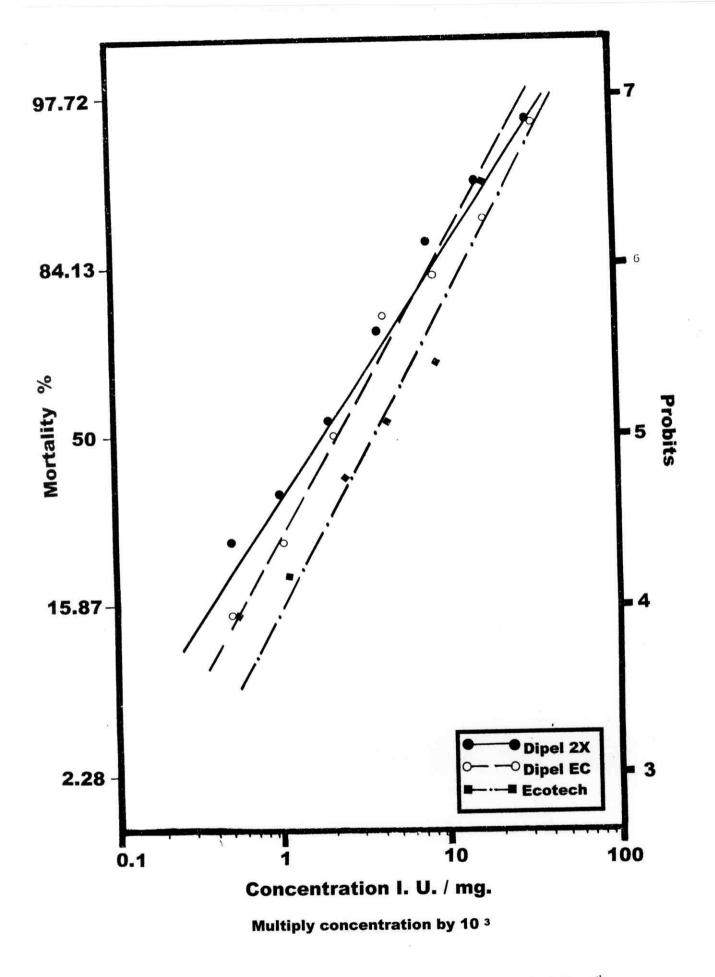


Fig. (5): Regression mortality lines due to feeding S. littoralis 4<sup>th</sup> instar larvae on castor-bean leaves treated with Dipel 2X, Dipel EC and Ecotech Bio.

## 1.1.f. Against 5th instar larvae :

As presented in Table (1), when  $5^{th}$  instar larvae of *S. littoralis* were fed on treated castor - bean leaves the mortality percentages recorded 13.3 - 93.3 % by Dipel 2X treatment at  $0.62 \times 10^3$  and  $40 \times 10^3$  IU/mg; 13.3 - 90.0 % (for those fed on leaves treated by Dipel EC at concentrations  $0.65 \times 10^3$  and  $41.6 \times 10^3$ ); and 16.7 - 93.3 % amongst those treated by Ecotech at concentration,  $0.7 \times 10^3 - 45 \times 10^3$  IU/mg.

As for the LC<sub>50</sub>'s and confidence limites after 5 days of treatments by the mentioned bioinsecticides; those were estimated by  $4.144 \times 10^3 (3.014 \times 10^3 - 5.616 \times 10^3)$ ,  $4.691 (3.400 \times 10^3 - 6.445 \times 10^3)$  and  $4.694 (3.379 \times 10^3 - 6.437 \times 10^3)$  IU/ mg. for Dipel 2X , Dipe EC and Ecotech, respectively (Table, 2 and Fig. 6).

# 1.1.g. Sixth instar larvae fed on treated food :

From data in Table (1), it is clear that the recorded corrected mortality percentages were 10.0- 90.0% (by feeding on castor – bean leaf discs treated with 0.75 x  $10^3$  –  $48.0 \times 10^3$  IU / mg. of Dipel 2X ), 6.7 – 86.7% ( $0.87 \times 10^3$  –  $56.0 \times 10^3$  IU /mg of Dipel EC ) and 10.0 – 90.0% ( $0.98 \times 10^3$  –  $63.0 \times 10^3$  IU/mg of Ecotech). While, data in Table (2) and those illustrated in Fig (7) indicate that the LC<sub>50</sub>'s after 5 days of treatment were  $5.939 \times 10^3$  IU/mg (with the confidence limites of  $4.288 \times 10^3$  –  $8.218 \times 10^3$ ),  $8.477 \times 10^3$  ( $6.133 \times 10^3$  –  $11.878 \times 10^3$  IU/mg) and  $7.574 \times 10^3$  IU /mg ( with the confidence limites of  $5.409 \times 10^3$  –  $10.575 \times 10^3$  IU/mg ) for Dipel 2X , Dipel EC and Ecotech, respectively.

The present results concerning the effect of *S. littoralis* larval feeding on castor — bean leaf discs treated with *B. thuringiensis* indicated that larval mortality increased with increasing the concentration of bioinsecticide, on one hand, and by treatment of larvae at their earlier instars, on the other hand.

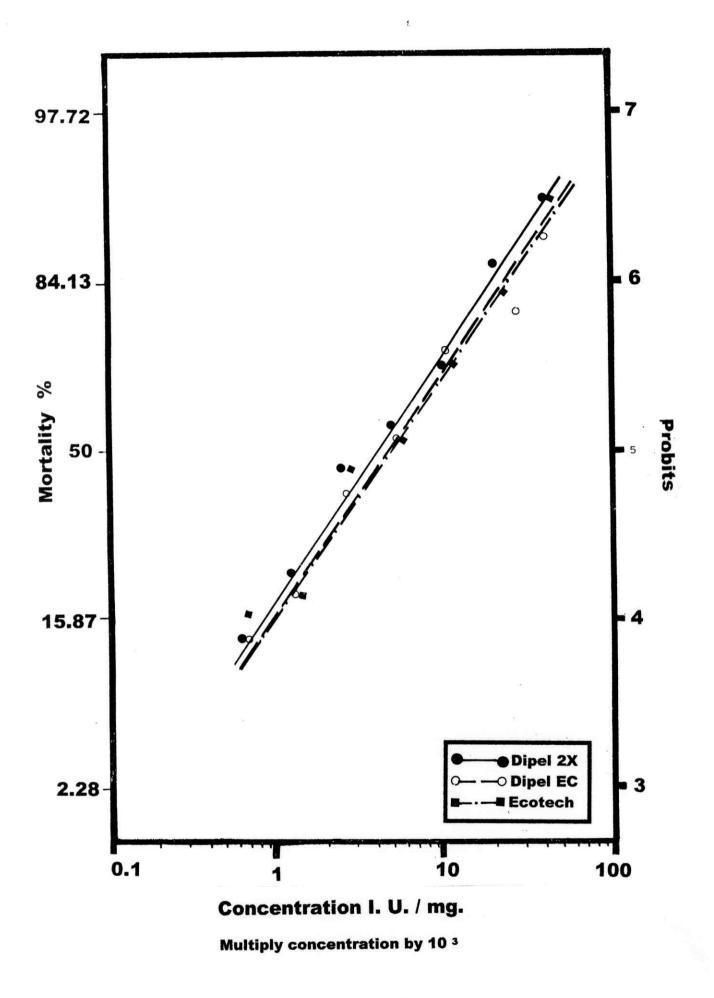


Fig. (6): Regression lines indicating mortality percentages among S. littoralis larvae fed in their 5<sup>th</sup> instar on treated castor - bean leaves treated with three bioinsecticides.

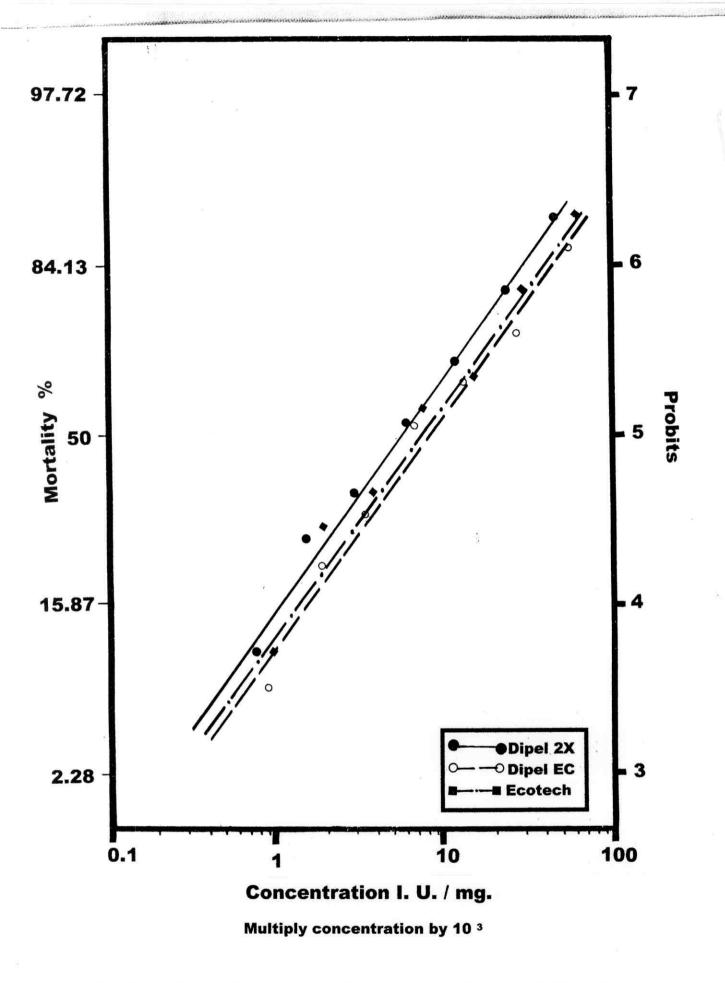


Fig. (7): Regression mortality lines after feeding of *S. littoralis*6 th instar larvae on castor-bean leaves treated with Dipel 2X,
Dipel EC and Ecotech Bio

. .

In this respect, Salama et al (1981) found that treatment of S. littoralis 250 and 500 ug / ml. diet caused 100% mortality, 12 - 17 days after exposure for 1, 2 or 3 days . Decreasing the dose to 125 or 200 ug / ml. Led to 100 %mortality after 17 - 22 days whether the exposure period was 1, 2, 3, 5or 7 days . Salama and Foda (1982) indicated that B. t. entomocidus showed a high potential activity, against S. littoralis, among 16 different varieties . Abdallah ( 1983 ) found that the highest concentration of 80.000 I U per mg of Thyricide HP and Bactospeine led to 82.5 and 86.6 % mortality, respectively when S. littoralis larvae were treated at their 2<sup>nd</sup> instar. Salama et al (1983) reported that B. t. kurstaki and B. t. aizawai were highly active against S. littoralis . Abou - Bakr et al (1993) studied the pathogenicity of 35 B. thuringiensis varieties against the 1st instar larvae of S. littoralis. They concluded that B. t. tolworthi (Hd - 301) was the most effective strain, causing 100% after 5 days of exposure, while three other strains mortality ( darmstadiensis , wushansis and aizawai ( HD-122 ) caused also 100% mortality, but after 7 days of exposure. Abdel - Halim (1997 a) found that the highest mortality percentages caused by Dipel 2X treatment to 2<sup>nd</sup> and 4<sup>th</sup> larval instars of S. littoralis occurred after 9 days and reached 95 and 72.1 %, respectively.

# 1.2. Efficacy on S. littoralis survivals after feeding of different larval instars on LC<sub>50</sub> of *B. thuringiensis* products:

Either of the six S. littoralis larval instars were fed on castor been leaves treated with the LC<sub>50</sub> of **B.** thuringiensis products for 48 hours after which the survivals were fed on clean castor bean leaves until pupation. The resultant adults from different treatments were caged and allowed to oviposit on Nerium oleander leaves. Different biological aspects were studied after treatment at each of the larval instars.

# 1.2.a. Treatment of newly hatched larvae:

As shown in Table (3), the  $LC_{50}$  of Dipel 2 X (0.464 x  $10^3$  IU/mg) caused the highest larval mortality percentage (58%), followed by Dipel EC (56%). While treatment by the LC50 of Ecotech (0.81 x  $10^3$  IU/mg) resulted 55% mortality among the treated larvae indicating that this preparation was the least effective on the larval stage.

# Larval period:

After feeding the freshly hatched S. littoralis larvae on the LC50's of Dipel 2X, Dipel EC and Ecotech, the surviving larvae after treatment showed significant prolongations in the larval period (16.8  $\pm$  0.8, 15.6  $\pm$ 0.9 and 15.4  $\pm$  1.0 days, respectively) than that of the control (12  $\pm$  0.4 days). Thus indicating that feeding of newly hatched larvae the LC50 of the bioinsecticides caused prolongations of the larval period. Statistical analysis of the obtained data indicated significant differences between the untreated and treated larvae. While, the differences in larval period by the three preparations were insignificant treatments between (Table, 3).

Table (3): Some biological parameters of S. littoralis stages after feeding of the newly hatched larvae on castor bean leaves treated with  $LC_{50}$  of three bioinsecticides (Results from treatment of 100 larvae).

Teatment			1.62	2.56				L.S.D. <sub>(0.05)</sub>
			9.26*	10.57*		×		F. (0.05)
Duration (in days)   Commitment			(7-9)	(11-13)				
% mortality among         Duration (in days)         % malformation (in days)         pupae	0.00	0.00	7.8 ± 0.4 a	12.0 ± 0.4 a	1.03	1.02	2.00	Control
% mortality among         Duration (in days)         % malformation (in days)<			(8-11)	(12-18)				$(0.8100 \times 10^3)$
% mortality among         Duration (in days)         % malformation (in days)         pupae         pupae         pupae         13.64         (14-18)         (10-13)         (10-13)         (10-13)         (10-13)         8.33         (14-19)         (9-11)         (14-19)         (14-19)         (14-19)         (14-19)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13)         (10-13) <th< td=""><td>8.57</td><td>10.25</td><td>9.2 ± 0.6 ac</td><td>15.4 ± 1.0 b</td><td>13.33</td><td>18.18</td><td>55.00</td><td>Ecotech</td></th<>	8.57	10.25	9.2 ± 0.6 ac	15.4 ± 1.0 b	13.33	18.18	55.00	Ecotech
% mortality among         Duration (in days)         % malformation (in days)			(9-11)	(14-19)				$(0.8896 \times 10^3)$
Sample   S	18.18	8.33	10.0 ± 0.3 bc	15.6 ± 0.9 b	18.18	21.43	56.00	Dipel EC
larvae prepupe pupae larvae** pupae pupae 58.00 24.14 9.09 16.8 ± 0.8 b 11.2 ± 0.3 b 13.64			(10-13)	(14-18)				$(0.4640 \times 10^3)$
larvae prepupe pupae larvae** pupae pupae pupae	15.00	13.64	$11.2 \pm 0.3$ b	$16.8 \pm 0.8^{\ b}$	9.09	24.14	58.00	Dipel 2X
% mortality among Duration (in days)	adult	pupae	pupae	larvae**	pupae	prepupe	larvae	(LU./mg)
	rmation	% malfo	(in days)	Duration	JQ.	6 mortality amon	0	Treatment

<sup>\*\*</sup> Larval period was estimated as the period from just after treatment until prepupa.

# Pre-pupal and pupal mortalities and pupal & adults malformations:

Amongst the prepupae that resulted from the *S. littoralis* surviving larvae, 24.14, 21.43 and 18.18% mortalities were recorded due to treatment of the freshly hatched larvae by the  $LC_{50}$ 's of Dipel 2X, Dipel EC and Ecotech, respectively, opposed to only 1.02% prepupal mortality in case of the control. The subsequent pupae suffered, also, mortalities that were estimated by 9.09, 18.18 and 13.33% for the three bacterial formulations, respectively. Amongst the obtained pupae, 13.64, 8.33 and 10.25% were found malformed (Fig. 8 A) after using Dipel 2X, Dipel EC and Ecotech, respectively, while 1.03% mortality and no malformation was recorded in case of the control pupae. After emergence, all the resultant adults from control were of normal shape, while 15, 18.18 and 8.57% of the adults (resulted after feeding the freshly hatched larvae on the  $LC_{50}$  of Dipel 2X, Dipel EC and Ecotech) were found malformed (Fig. 8 B).

#### Pupal period:

Significant elongations in the pupal period were also detected due to treatment of freshly hatched *S. littoralis* lravae by Dipel 2X or Dipel EC 11.2  $\pm$  0.3 and 10  $\pm$  0.3 days, respectively) and that of the control (7.8  $\pm$  0.4 days). While, the pupal period in case of Ecotech treatment (9.2  $\pm$  0.6 days) was insignificantly longer than control and insignificantly shorter than Dipel EC treatment (Table, 3).

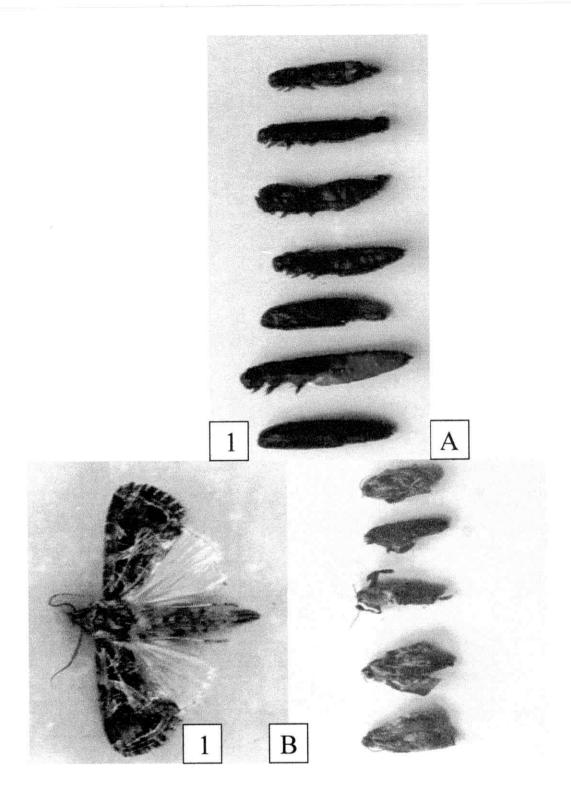


Fig. (8): Malformed S. littoralis stages due to feeding the larvae stage on castor – bean leaves treated with B. thuringiensis preparations.

- A. Normal (1) and malformed pupae.
- B. Normal (1) and malformed adults.

#### Fecundity and fertility of emerged moths:

Table (4) domonstrates the fecundity of the cotton leafworm moths after being treated as newly hatched larvae by the LC<sub>50</sub> of Dipel 2X, Dipel EC and Ecotech.

Data indicated that treatments caused reductions in the oviposition period, average number of deposited eggs / female and hatchability %. The average number of eggs per a control female was  $530.6 \pm 6.2$ . This number was reduced significantly to  $136.2 \pm 6.3$ ,  $143.0 \pm 5.5$  and  $156.6 \pm 1.8$  eggs/female due to treatment by the LC<sub>50</sub> values of Dipel 2X, Dipel EC and Ecotech, respectively. All reductions in the average number of deposited eggs/female due to biocidal treatment were significant compared to that recorded for the control treatment. The severest reduction was that occurred due to Dipel 2X treatment, although this value was insignificantly lower than those recorded after Dipel EC and Ecotech treatments.

## · Adults' langevity:

From data presented in Table (4), it could be deduced that feeding of freshly hatched *S. littoralis* larvae on bioinsecticide treated castor-bean leaves caused significant reductions in the longevities of subsequent male and female adults (5.2 & 5.6, 4.8 & 5.2 and 4.2 & 5.8 days for males and females after larval treatment by Dipel 2X, Dipel EC and Ecotech, respectively opposed to 8.6 & 9 days for the control adults. The severest reduction in male's longevity than control was that occurred by Ecotech treatment, while that of female occurred after larval treatment by Dipel EC, but, all the differences between treatments were insignificant. It could be generally observed that female's longevity was slightly longer than of that the male (Table, 4).

Table (4): Effect of S. littoralis freshly hatched larval treatment by B. thuringiensis preparations on fecundity and longevity of the resultant adults. (Results from 3 pairs / treatment ).

Treatment (I.U./mg).	Oviposition period (days)	Averag	Average no. of	%	Adult longevity (days) ± S.E.	ity (days)
	±S.E.	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	hatching	male	female
Dipel 2X	5.4 ± 0.5 b	136.2 ± 6.3 b	$77.8 \pm 6.4^{ b}$	57.12	$5.2 \pm 0.3^{\ b}$	5.6 ± 0.3 b
$(0.4640 \times 10^3)$	(4-7)	(120-151)	(60-94)		(3-7)	(3-7)
Dipel EC	$4.9 \pm 0.3^{\ b}$	$143.0 \pm 5.5^{b}$	$79.0 \pm 5.7^{\text{ b}}$	55.24	$4.8 \pm 0.4^{\ b}$	5.2 ± 0.4 b
$(0.8896 \times 10^3)$	(4-6)	(130-160)	(70-100)		(3-7)	(3-7)
Ecotech	$5.0 \pm 0.3^{\ b}$	156.6 ± 1.8 b	110.0 ± 3.6 °	70.24	$4.2 \pm 0.2^{\text{ b}}$	5.8 v 0.4 b
$(0.8100 \times 10^3)$	(4-6)	(150 - 162)	(96-116)		(3-5)	(3-7)
Control	8.8 ± 0.4 <sup>a</sup>	530.6 ± 6.2 a	490.2 ± 5.7 °a	92.38	8.6 ± 0.1 a	9.0 ± 0.1 a
	(8-10)	(506 - 596)	(470-500)		(8-10)	(8-10)
F. (0.05)	22.8*	446.31*	508.58*		21.46*	18.17*
L.S.D. (0.05)	1.34	32.30	13.84		1.47	1.40

#### 1.2.b. Treatment of the 24 h. old 1st instar larvae:

After feeding of the 24 h. old  $1^{st}$  instar larvae of *S. littoralis* on the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech, the mortality percentages were estimated among the larvae and subsequent stages. Among the treated larvae, the highest mortality percentages (54.67%) was recorded from Dipel 2X treatment followed by Dipel EC and Ecotech (54.00%) (Table, 5). Results indicated that the three bioinsecticidal preparations were, nearly, of similar effect on larval mortality % when treatment took place on first instar larvae (Table, 5).

#### Larval period:

Data presented in Table (5), clearly, indicate that treatment of *S*. *littoralis* 1<sup>st</sup> instar larvae with any of the three tesed bioinsecticides caused significant prolongations in the larval period than control.

The shortest larval period  $(11.4 \pm 0.3 \text{ days})$  was recorded from the control larvae. This period was significantly prolonged to  $16.2 \pm 1.0$ ,  $15.4 \pm 0.7$  and  $15.6 \pm 0.9$  days by 1<sup>st</sup> instar larval treatment on Dipel 2X, Dipel EC and Ecotech, respectively. Accordingly, the severest prolongation on larval period was that of Dipel 2X, but the differences were insignificant, compared with Dipel EC and Ecotech (Table, 5).

# Prepupal and pupal mortalities and pupal & adults' malformations:

Data in Table (5) show, that Dipel 2X caused the highest mortality percentage of prepupae (25.00%), opposed to 23.19, 17.39 and 1.02% in cases of Dipel Ec and Ecotech treatment and control. Amongst the obtained pupae, the highest mortality percentage (19.6%) occurred also due to Dipel 2X treatments followed by 10.53% from Ecotech treatment and 9.43% from Dipel EC treatment. Also, highest percentage of

Table (5): Some biological parameters of S. littoralis stages after feeding of the 24h. old 1st larval instar on castor bean leaves treated with  $LC_{50}$  of three bioinsecticides (Results from treatment of 100 larvae / treatment ).

Treatment	0	% mortality among	94	Duration (in days)	(in days)	% malformation	rmation
(LU/mg)	larvae	prepupe	pupae	larvae	pupae	pupae	adult
Dipel 2X	54.67	25.00	19.61	$16.2 \pm 1.0^{\ b}$	$10.2 \pm 0.7^{\ b}$	21.57	17.65
$(0.9984 \times 10^3)$				(14-20)	(9-11)		
Dipel EC	54.00	23.19	9.43	$15.4 \pm 0.7$ b	$9.0 \pm 0.8$ bc	15.91	16.22
$(1.2256 \times 10^3)$				(13-18)	(8-10)		
Ecotech	54.00	17.39	10.53	$15.6 \pm 0.9^{\ b}$	8.2 ± 1.6 ac	13.15	15.15
$(1.1880 \times 10^3)$				(13-18)	(6-9)		
Control	2.00	1.02	1.03	$11.4 \pm 0.3$ a	7.4 ± 0.2 a	0.00	0.00
				(11-12)	(7-8)		
F-(0.05)				10.47*	7.50*		
L.S.D. <sub>(0.05)</sub>				2.34	1.48		

malformed pupa (21.57%) occurred after first instar larval treatment by Dipel 2X, while the lowest percentage (13.15%) was recorded by 1<sup>st</sup> instar larval feeding on castor bean leaves treated with the LC<sub>50</sub> of Ecotech. Dipel EC treatment caused intermediate effect (15.9% malformed pupae). Among the subsequent moths, certain malformations could be detected. Those reached 17.65, 16.22 and 15.15% when the 1<sup>st</sup> instart larvae were fed on Dipel 2X, Dipel Ec and Ecotech, respectively (Table, 5).

#### Pupal period:

As shown in Table (5), insignificant elongation in the pupal period occurred due to  $1^{st}$  instar larval treatment by Ecotech (8.2  $\pm$  1.6 days) than that occurred from the control pupae (7.4  $\pm$  0.3 days). While, this period became significantly longer than control due to Dipel EC (9  $\pm$  0.8 days) and Dipel 2X (10.2  $\pm$  0.7 days) treatments. The severest prolongation in pupal period occurred due to Dipel 2X treatment, but the effect was insignificant compared to Dipel EC treatment and significant compared to Ecotech treatment (Table, 5).

# · Fecundity and fertility of emerged moths:

Results in Table (6) indicated that fecundity and fertility of moths resulted from treatment of  $1^{st}$  instar larvae with LC<sub>50</sub> of either of the three bio-insecticides were, significantly reduced. There were reductions in the oviposition period, average number of eggs/female and hatchability %. The average oviposition period of a control moth female reached  $8.3 \pm 0.4$  days, and this period was found to be reduced to 5, 5.1 and 5.4 days due to larval treatment by Dipel 2X, Dipel EC and Ecotech, respectively. Throughout the oviposition period, a fertilized female from the three

Table (6): Effect of S. littoralis 1st instar larval treatment by B. thuringienis preparations on fecundity and longevity of the resultant adults (Results from 3 pairs).

			11014	11714	13 64*	5
(0.10)	(07)					
(8-10)	(8-9)		(470 - 500)	(496 - 530)	(8-10)	
9.0 ± 0.2 °	8.6 ± 0.1 *	93.63	488.2 ± 5.7 °	314.6 ± 6.2	8.3 ± 0.4	Control
			600	712/-/00	03-04	
(3-7)	(3-5)		(117-125)	(162-170)	(5-7)	(1.880 x 10 <sup>3</sup> )
5.8 ± 0.4 b	4.2 ± 0.2 b	72.03	120.0 ± 1.0 °	166.6 ± 1.6 °	5.4 ± 0.5 °	Ecotech
(3-7)	(3-7)		(80-95)	(133-171)	(4-7)	(1.2256 x 10°)
5.2 ± 0.3 °	5.0 ± 0.4 °	56.65	86.0 ± 3.0 "	151.8 ± 6.3 55	5.1 ± 0.5	Dipel EC
(3-1)	(3-1)		(/0-94)	(101-001)	(+-/)	(0.20t A 10)
3	(2)		(70.07)	(130 161)	(4-7)	(0 9984 v 10 <sup>3</sup> )
5.6 ± 0.3 b	$5.0 \pm 0.3^{\ b}$	56.86	82.0 ± 4.5 b	144.2 ± 5.5 b	5.0 ± 0.5 b	Dipel 2X
				± S.E.		
			± S.E.	female		
female	male	hatching	hatched larvae	deposited eggs/	+ S.E.	
E.	± S.E.	%			period (days)	(1.U./mg).
vity (days)	Adult longevity (days)		Average no. of	Averag	Oviposition	Treatment

treatments deposited  $82 \pm 4.5$ ,  $86 \pm 3.0$  and  $120 \pm 10$  eggs, respectively, oviposed to  $514.6 \pm 6.2$  eggs / a control female. Among the deposited eggs, the hatchability percentages were 56.86, 56.65, 72.03 and 93.63, respectively. Thus indicating severe effect of larval feeding on Dipel 2X or Dipel EC treated castor-bean leaves on the fecundity of resultant moths and fertility of deposited eggs. This effect was more detectable than those recorded from Ecotech treatment, although in all cases the effect of the three bioinsecticidal preparations was significant, compared to results from control females (Table, 6).

# Adults' longevity:

The effect of *S. littoralis* 1<sup>st</sup> instar larval feeding on bioinsecticide treated castor-bean leaves was also clear on the longevity of male and female moths which showed, significantly, shorter life-span than control. In case of adults resulted from larvae treated by Dipel 2X, Dipel EC and Ecotech, the recorded longevities were 5 & 5.6, 5 & 5.2 and 4.2 & 5.8 days for male & female, respectively, opposed to 8.6 & 9 days in case of the control moths (Table, 6). It could be also observed from the same Table that the differences between moth longevities from the three treatments were insignificant.

## 1.2.c. Treatment of 2<sup>nd</sup> instar larvae:

The efficacy of the three bioinsecticidal preparations under study on S. *littoralis* was assayed when offered to the second instar larvae castor-bean leaves treated with the LC<sub>50</sub>'s of the these compounds.

The highest mortality percentage amongst the treated larvae (58.5%) occurred from Dipel EC treatment (Table, 7). That was followed by Dipel 2X (57%) and Ecotech (54%).

Table (7): Some biological parameters of S. littoralis stages after feeding of the 2<sup>nd</sup> larval instar on treated castor bean leaves treated with LC<sub>50</sub> of three bioinsecticides (Results from treatment of 100 larvae).

Treatment	0	% mortality among	90	Duration (in days)	(in days)	% malformation	mation
(I.U./mg)	larvae	prepupe	pupae	larvae	pupae	pupae	adult
Dipel 2X	57.00	22.64	7.32	14.8 ± 0.6 °	$10.2 \pm 0.5$ b	13.16	27.27
$(1.0592 \times 10^3)$				(13-16)	(9-11)		
Dipel EC	58.50	25.30	4.84	$13.0 \pm 0.7^{\ b}$	$10.0 \pm 0.5$ bc	18.96	25.00
$(1.5181 \times 10^3)$				(11-15)	(9-11)		
Ecotech	54.00	19.56	8.11	12.2 ± 0.4 b	$8.4\pm0.8$ ac	11.43	16.13
$(1.3518 \times 10^3)$				(11-13)	(7-11)		
Control	2.00	1.02	1.03	10.3 ± 0.4 a	7.4 ± 0.3 a	0.00	0.00
				(9-11)	(7-9)		
F <sub>*(0.05)</sub>				4.79*	6.77*		
L.S.D. (0.05)				1.76	1.67		

#### Larval period:

Data presented in Table (7), clearly, indicate that treatment of S. *littoralis*  $2^{nd}$  larval instar with any of the three tested bioinsecticides caused significant prolongation in the larval period than the control. The shortest larval period  $10.3 \pm 0.4$  days was that of the control. This period was prolonged to  $12.2 \pm 0.4$ ,  $13.0 \pm 0.7$  and  $14.8 \pm 0.6$  days due to  $2^{nd}$  instar larval treatment by Ecotech, Dipel EC and Dipel 2X, respectively indicating the severest effect of Dipel 2X which caused significant prolongation in the larval period than Dipel EC and Ecotech (Table, 7). While the difference in larval period was insignificant between Dipel Ec and Ecotech, although remained, significantly, longer than that of the control (Table, 7).

# Prepupal and pupal mortalities and pupal & adults malformations:

As shown in Table (7), feeding of the 2<sup>nd</sup> instar larvae on castor-bean leaves treated by the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech caused 22.64, 25.3 and 19.56 % mortalities among the subsequent prepupae, and 7.32, 4.84 and 8.11% mortalities among the resultant pupae. Thus indicating that the severest effect on prepupae occurred by Dipel EC treatment, followed by Dipel 2X, while on pupae, the severest effect occurred due to Ecotech followed by Dipel 2X larval treatment.

As for the observed malformations among the obtained pupae and adults, those reached 13.16, 18.96 and 11.43% of pupae and 27.23, 25 and 16.13% of adults by larval treatment by Dipel 2X, Dipel EC and Ecotech, respectively, indicating highest effect on pupae by Dipel EC and that on adults by Dipel 2X (Table, 7).

#### Pupal period:

Larval treatments in the  $2^{nd}$  instar caused, also, prolongations in the larval period than that recorded from those resulted from the control pupae (7.4  $\pm$  0.3 days). As shown in Table (7), this prolongation was insignificantly longer than that recorded from the control in case of Ecotech treatment (8.4  $\pm$  0.8 days), but significantly longer than control in cases of Dipel EC and Dipel 2X (10  $\pm$  0.5 and 10.2  $\pm$  0.5 days, respectively; Table, 7).

# · Fecundity and fertility of emerged moths:

Table (8) shows, the reductions in oviposition period, average number of deposited eggs/ female and hatching percentages of S. *littoralis* due to  $2^{nd}$  larval instar treatments by B. *thuringiensis* products. All reductions were significant compared to the control treatment. While, the differences between treatments were insignificant. Generally, the severest effect was that of Dipel 2X treatment which produced shortest oviposition period  $(4.7 \pm 0.4 \text{ days})$ , fewest number of eggs/female  $(178.6 \pm 8.7 \text{ eggs})$  and the lowest hatchability % (55.99%), Dipel EC came the next (5.3 days as oviposition period, 179 deposited eggs/female and 56.42% as hatchability %). While, the lowest effect among the three preparations, occurred due to Ecotech treatment 5.3 days, 199.7 eggs and 70.1%; these values were insignificantly higher than those of Dipel 2X and Dipel EC and significantly lower than those recorded from the control check (9 days, 526.2 eggs/ female) and 93.12% hatching; Table, 8).

## · Adults' longevity:

As presented in Table (8), the longevities of S. littoralis adults

Table (8): Effect of S. littoralis 2<sup>nd</sup> instar larval treatment by B. thuringiensis preparations on fecundity and longevity of the resultant adults (Result from 3 pairs).

Treatment (I.U./mg).	Oviposition period (days)	Averag	Average no. of	%	Adult longevity (days) $\pm$ S.E.	vity (days) E.
	± S.E.	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	hatching	male	female
Dipel 2X	4.7 ± 0.4 b	178.6 ± 8.7 b	100.0 ± 9.3 °	55.99	4.6 ± 0.3 b	4.8 ± 0.3 b
$(1.0592 \times 10^3)$	(4-6)	(155-200)	(70-120)		(3-6)	(3-6)
Dipel EC	5.3 ± 0.5 b	$179.0 \pm 9.7^{b}$	101.0 ± 0.3 °	56.42	$5.6 \pm 0.1^{\ b}$	5.6 ± 0.1 b
$(1.5184 \times 10^3)$	(4-7)	(153-208)	(100 - 102)		(5-6)	(5-6)
Ecotech	5.3 ± 0.3 b	$199.7 \pm 2.4^{b}$	140.0 ± 6.4 b	70.10	$5.0 \pm 0.1$ b	5.6 ± 0.1 b
$(1.3518 \times 10^3)$	(4-6)	(193-208)	(120-160)		(4-6)	(5-6)
Control	$9.0 \pm 0.4^{a}$	526.2 ± 13.8 a	490.0 ± 6.4 <sup>a</sup>	93.12	8.6 ± 0.1 a	9.0 ± 0.1 a
	(8-10)	(494-574)	(470-510)		(8-9)	(8-10)
F. (0.05)	36.91*	179.94*	191.09*		20.06*	18.06*
L.S.D. (0.05)	1.37	32.69	20.33		1.39	1.52

previously treated as 2<sup>nd</sup> instar larvae with bioinsecticides were significantly shorter, in cases of males and females, than those obtained from the control moths. Longevities of males were 4.6, 5.6 and 5 days for 2<sup>nd</sup> instar larval treatment by Dipel 2X, Dipel EC and Ecotech, respectively, while those of females were 4.8, 5.6 and 5.6 days, respectively, opposed to 8.6 and 9 days for male and female of the control moths.

# 1.2.d. Third instar larval treatments:

As shown in Table (9), feeding the *S. littoralis*  $3^{rd}$  instar larvae on castor-bean leaves treated with the LC<sub>50</sub> of either of the three products of *B. thuringiensis*; Dipel 2X, Dipel EC and Ecotech caused 56, 52 and 54% mortalities amongst the treated larvae, opposed to only 2% mortality among larvae of the control check.

#### Larval period:

As occurred with the two former instar, treatment of *S. littoralis*  $3^{rd}$  instar larvae caused significant prolongations in the larval period that reached a maximum of  $13.4 \pm 0.5$  days by Dipel 2X treatment, the period which was insignificantly longer than Dipel EC treatment ( $12.8 \pm 0.4$  days) and significantly longer than Ecotech treatment ( $11.6 \pm 0.5$  days) and the untreated larvae ( $8.6 \pm 0.3$  days; Table, 9). Statistical analysis showed also that the difference in larval period between Dipel EC and Ecotech treatments was insignificant (Table, 9).

Table (9): Some biological parameters of S. littoralis stages after feeding of the 3<sup>rd</sup> larval instar on castor bean leaves treated with LC<sub>50</sub> of three bioinsecticides (Results from treatment of 100 larvae).

		2.11	1.26				L.S.D. (0.05)
		4.49*	34.03*				F-(0.05)
		(6-9)	(8-9)				
0.00	0.00	7.5 ± 0.5 <sup>a</sup>	8.6 ± 0.3 a	0.67	0.67	2.00	Control
		(7-13)	(10-13)				(2.0772 x 10°)
13.33	11.76	9.2 ± 1.2 ab	11.6 ± 0.5 b	10.53	17.39	54.00	Ecotech
		(9-10)	(12-14)				(2.0112 x 10°)
22.73	18.92	9.8 ± 0.4 b	$12.8 \pm 0.4$ bc	9.75	21.15	52.00	Dipel EC
		(8-12)	(13-14)				(1.6224 x 10°)
25.00	11.11	$10.0 \pm 0.7^{\ b}$	13.4 ± 0.5 °	18.18	17.86	56.00	Dipel 2X
adult	pupae	pupae	larvae	pupae	prepupe	larvae	(1.U./mg)
% malformation	% malf	Duration (in days)	Duration		% mortality among		Treatment

# Pre-pupal & pupal mortalities and pupal & adult's malformations:

By feeding the  $3^{rd}$  instar *S. littoralis* larvae on castor-bean leaves contaminated by the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech, the percentages of 17.86, 21.15 and 17.39% mortalities were recorded, respectively among the subsequent pupae, while those occurred among the pupae were 18.18, 9.75 and 10.53%, respectively. Thus indicating the severest effect on both stages by Dipel 2X product. The same preparation caused, also, the highest percentage of adults' malformation (25%), while Dipel EC caused the highest % of pupal malformation (18.92%, Table 9).

#### Pupal period:

Among the successful *S. littoralis* pupae that resulted from  $3^{rd}$  instar larval treatment by bioinsecticides (81.82 – 90.25%), the pupal period showed also prolongations compared to that of the control pupae (7.5 ± 0.5 days). These periods were significantly longer than control in cases of Dipel 2X and Dipel EC treatments (10 ± 0.7 and 9.8 ± 0.4 days, respectively; Table, 9) and insignificantly longer than control after Ecotech treatment (9.2 ± 1.2 days).

# Fecundity and fertility of emerged moths:

As shown in Table (10), feeding the  $3^{rd}$  instar larvae of *S. littoralis* on castor-bean leaves treated with the LC<sub>50</sub> of Dipel 2X led to female moths of significantly, shorter oviposition periods  $(4.9 \pm 0.3, 5.2 \pm 0.4$  and  $5 \pm 0.5$  days, respectively) than that recorded from resulted from the untreated larvae  $(8.44 \pm 0.4 \text{ days})$ . Female moths from the treated larvae deposited, significantly, fewer number of eggs  $(221 \pm 9.51, 228.8 \pm 10.0)$ 

Table (10): Effect of S. littoralis 3rd larval instar treatment by B. thuringiensis perparations on fecundity and longevity of the resultant adults (Results from 3 pairs).

Treatment (I.U./mg).	Oviposition period (days)	Averag	Average no. of	%	Adult longevity (days)	ity (days)
	± S.E.	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	hatching	male	female
Dipel 2X	4.9 ± 0.3 b	221.0 ± 9.5 °	130 ± 10.8 °	58.82	46+04b	5/1+02b
$(1.6224 \times 10^3)$	(4-6)	(170-270)	(95-190)		(3-7)	0.7 1 0.5
Dipel EC	5.2 ± 0.4 b	228.8 ± 10.0 °	152.4 ± 8.3 °	68.40	48+04b	50+01b
$(2.0112 \times 10^3)$	(4-7)	(195-250)	(141 - 180)		(3-7)	(3-7)
Ecotech	5.0 ± 0.5 b	295.0 ± 6.4 b	212.0 ± 11.4 b	71.86	52+04b	56+036
$(2.0772 \times 10^3)$	(4-7)	(280 - 315)	(190 - 250)		(3-7)	(3-7)
Control	8.4 ± 0.4 <sup>a</sup>	534.4 ± 5.3 <sup>a</sup>	498.2 ± 5.7 °	93.63	8.6 ± 0.1 a	9.0 ± 0.1 a
	(8-10)	(518 - 550)	(490 - 511)		(8-9)	(8-10)
F. (0.05)	24.58*	322.67*	1028.17*		23.02*	18.81*
L.S.D. (0.05)	1.17	38.06	42.37		1.35	1.49

and  $295 \pm 6.4$  eggs/female, respectively) than control female ( $534.4 \pm 5.3$  eggs). Also , the hatching percentages were lower among eggs from treated females than control (58.82, 68.4 and 71.86%, respectively, opposed to 93.63% among eggs from the control females). These data indicated that the severest effect occurred by Dipel 2X treatment which resulted in female moths showed oviposition period and deposited fewest number of eggs/female through which the lowest hatchability % occurred). The recorded average number of eggs/female due to Dipel 2X was statistically insignificant compared to those obtained after Dipel EC treatment and significantly higher than those deposited by a single female resulted from larval treatment by Ecotech (Table, 10).

#### Adults' longevity:

Insignificant differences were recorded in the males and females longevities among moths resulted from larvae treated with Dipel 2X, Dipel EC and Ecotech (4.6 & 5.4, 4.8 & 5 and 5.2 & 5.6 days for males and females, respectively; Table, 10). But in all cases, the mentioned life-spans were, significantly, shorter than those recorded for males and females resulted from the control check (8.6 and 9 days, respectively).

# 1.2.e. Treatment of 4<sup>th</sup> larval instar:

From data in Table (11), it is clear that the highest mortality percentage occurred when larvae were fed on castor-bean leaves contaminated with the  $LC_{50}$  of Dipel 2X (56%), followed by Ecotech (55%) and Dipel EC (54%).

## Larval period:

From data tabulated in Table (11), it is clear that Dipel 2X treatment to the 4<sup>th</sup> instar larvae caused, statistically, longer larval period

Table (11): Some biological parameters of S. littoralis stages after feeding of the 4th larval instar on treated castor-bean leaves with LC<sub>50</sub> of three bioinseciticdes (Results from treatment of 100 larvae).

		N.S.	2.18				L.S.D. (0.05)
		2.51	3.55*				F.(0.05)
		(7-8)	(9-10)				
0.00	0.00	$7.4 \pm 0.4$	9.4 ± 0.3 a	1.04	2.04	2.00	Control
		(7-10)	(8-13)				$(2.6540 \times 10^{\circ})$
13.33	14.28	$8.4 \pm 0.5$	$10.2 \pm 0.9^{\text{ ab}}$	7.89	15.55	55.00	Ecotech
		(7-10)	(9-13)			(E) (1) (3)	$(2.2672 \times 10^{\circ})$
10.34	17.14	9.0± 0.6	11.0± 0.7 ab	7.89	17.39	54.00	Dipel EC
		(8-10)	(11-14)				$(1.8432 \times 10^{\circ})$
12.5	25.00	$9.2 \pm 0.5$	12.2 ± 0.6 b	8.57	20.45	56.00	Dipel 2X
adult	pupae	pupae	larvae	pupae	prepupe	larvae	(LU/mg)
rmation	% malformation	(in days)	Duration (in days)	54	% mortality among	,	Treatment

 $(12.2 \pm 0.6 \text{ days})$  than control  $(9.4 \pm 0.3 \text{ days})$ . While, larval treatments by the LC<sub>50</sub> of either Dipel EC or Ecotech led to insignificant elongations in the larval period  $(11 \pm 0.7 \text{ and } 10.2 \pm 0.9 \text{ days})$  than control, although these periods remained insignificantly shorter than that recorded by Dipel 2X treatment (Table, 11).

# Prepupal & pupal mortality and pupal and adults malformations:

By using the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech for castorbean treatments to be offered as food to the *S. littoralis*  $4^{th}$  instar larvae, the subsequent prepupae & pupae showed the mortality percentages of 20.45 & 8.57, 17.39 & 7.89 and 15.55 & 7.89%, respectively (Table, 11).

While, among the resultant pupae & adults, the percentages of 25 & 12.5, 17.14 & 10.34 and 14.28 & 13.33% malformed individuals were, respectively, detected (Table, 11). Thus indicating severest effect on prepupal and pupal mortalities and pupal malformations due to Dipel 2X treatment and adults' malformations by Ecotech treatment.

### Pupal period:

Analysis of variance between pupal periods of treated and untreated larvae indicated that there was no-significant difference between pupal period of the control check (7.4 days) and those recorded from Dipel 2X, Dipel EC and Ecotech (9.2, 9 and 8.4 days, respectively; Table, 11).

#### Fecundity and fertility of emerged moths:

As occurred with the aforementioned larval instars, when the treated castor - bean leaves were offered to the freshly moulted 4<sup>th</sup> instar larvae, that caused significant reductions in the average numbers of deposited eggs/female than those counted from a single mated female from the control check ( $283.8 \pm 5.8$ ,  $286 \pm 4.9$  and  $297 \pm 0.9$  eggs after larval treatment by the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech, respectively, opposed to  $540 \pm 6.4$  eggs/female by a control female, Table, 12). While, the differences in average numbers of deposited eggs/female from treated larvae were, statistically, insignificant.

#### · Adults' longevity:

As shown in Table (12), treatments of the 4<sup>th</sup> larval instar with either of the three bioinsecticides caused significant reductions in the male and female adults' longevity which reached 4.6 & 6.6, 4.2 & 4.6 and 5 & 5.8 days due to treatment by Dipel 2X, Dipel EC and Ecotech, respectively, opposed to 8.8 & 9.4 days as life-spans of the control check moths. While, on the other hand, the differences in average life-span between males and females from the three treatments were, statistically, insignificant, although the shortest life-spans of both sexes were recorded from Dipel EC treatment (Table, 12).

### 1.2.f. Treatment of S. littoralis 5th larval instar:

From data in Table (13), it is clear that the highest larval mortality percentage occurred when larvae were fed on castor-bean leaves contaminated with the LC<sub>50</sub> of Dipel 2 X (55%), followed by Dipel EC and Ecotech (54%).

Table (12): Effect of S. littoralis 4th larval instar treatment by B. thuringiensis preparations on fecundity and longevity of the resultant adults (Results from 3 pairs).

Treatment (I.U./mg).	Oviposition period (days)	Averag	Average no. of	%	Adult longevity (days) $\pm$ S.E.	⁄ity (days) €.
	± S.E.	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	hatching	male	female
Dipel 2X	4.2 ± 0.4 b	283.8 ± 5.8 b	200.0 ± 9.1 °	70.47	4.6 ± 0.2 b	6.6 ± 0.1 b
$(1.8432 \times 10^3)$	(3-5)	(270-300)	(18-230)		(3-5)	(5-7)
Dipel EC	$4.4 \pm 0.5^{\ b}$	286.0 ± 4.9 b	202.0 ± 5.6 °	70.63	4.2 ± 0.4 b	4.6 ± 0.5 b
$(2.2672 \times 10^3)$	(3-6)	(272 –300)	(188-220)		(3-7)	(3-7)
Ecotech	$5.2 \pm 0.4^{\ b}$	297.0 ± 0.9 b	230.0 ± 5.6 b	77.44	$5.0 \pm 0.3^{\ b}$	5.8 ± 0.2 b
$(3.6540 \times 10^3)$	(4-6)	(296 –300)	(220-250)		(3-7)	(5-7)
Control	$8.7 \pm 0.4^{8}$	540.0 ± 6.4 <sup>a</sup>	501.0 ± 2.9 a	92.78	8.8 ± 0.1 <sup>8</sup>	9.4 ± 0.2 °
	(8-10)	(520-560)	(492-501)		(8-9)	(9-11)
F. (0.05)	23.85*	574.14*	469.15*		21.54*	19.25*
L.S.D. (0.05)	1.48	17.98	23.20		1.57	1.78

#### Larval period:

Table (13) shows that the larval period of the surviving larvae after Dipel 2X and Dipel EC treatments at the LC<sub>50</sub> levels showed significant prolongations from a mean of 7.8 days for the control larvae and also for those fed in their 5<sup>th</sup> instar on Ecotech treated castor-be leaves to 9.4  $\pm$  0.7 in case of Dipel 2X treatment , and insignificant prolongation to 9  $\pm$  0.7 days by Ecotec treatment .

### Prepupal & pupal mortality and pupal and adults' malformations:

As shown in Table (13), the prepupal and pupal mortality percentages were higher by 5<sup>th</sup> instar larval treatments by Dipel EC (15.38 & 20.45%) and Dipel 2X (13.33 & 20.51%) than Ecotech (9.84 & 19.65 %). The highest percentage of malformed pupae (14.28%) occurred by feeding on Dipel EC and that of malformed adults (22.22%) by feeding on Dipel 2X (Table, 13).

#### · Pupal period:

Analysis of variance between pupal period of those resulted from treated and untreated larvae indicated that there was no-significant difference, although the pupal periods for those resulted after 5<sup>th</sup> instar larval treatment were longer than control (7.4 days; Table, 13).

## Fecundity and fertility of emerged moths:

Results indicated significant reductions in longevity, fecundity and fertility of *S. littoralis* moths emerged after different larval treatments than control (Table, 14). The oviposition period lasted  $8.7 \pm 0.4$  days for the control female moths; this period was reduced to  $5.9 \pm 0.1$ ,  $6 \pm 0.3$ 

Table (13): Some biological parameters of S. littoralis stages after feeding of the 5th larval instar on castor-bean leaves treated with  $LC_{50}$  of three bioinseciticdes (Results from treatment of 100 larvae).

Treatment		% mortality among	20	Duration	Duration (in days)	% malformation	rmation
(LU/mg)	larvae	prepupe	pupae	larvae	pupae	pupae	adult
Dipel 2X	55.00	13.33	20.51	9.4 ± 0.7 b	82+06	12 90	22.22
(4140					i		44.44
(4.1440 x 10°)				(8-12)	(7-10)		
Dipel EC	54.00	15.38	20.45	$9.00 \pm 0.7$ ab	$8.2 \pm 0.4$	14.28	16.67
$(4.6912 \times 10^3)$				(7-11)	(7-9)		
Ecotech	54.00	9.84	19.65	7.8 ± 0.7 a	8.0 ± 0.6	6.67	14 28
$(4.6944 \times 10^3)$				(6-10)	(7-10)		
Control	2.00	1.02	0.00	7.8 ± 0.4 °	$7.4\pm0.5$	0.00	0.00
				(7-9)	(7-9)		
F. <sub>(0.05)</sub>				4.93*	0.45		
L.S.D. (0.05)				1.28	N.S.		

and  $6.2 \pm 0.4$  days after 5<sup>th</sup> instar larval treatment by Dipel 2X, Dipel EC and Ecotech. Also, the average of deposited eggs averaged  $530 \pm 13.6$  eggs/ a control female, and average was found to be reduced to  $368 \pm 10.2$ ,  $380 \pm 7.2$  and  $390 \pm 7.2$  eggs/ female by using the aforementioned bioinsecticides, respectively. From these eggs the % hatching reached 76.09, 78.95 and 82.05 % respectively, opposed to 91.13% among eggs deposited by the control females (Table, 14).

#### · Adults' longevity:

Table (14) shows the longevities of *S. littoralis* male and female adults treated as larvae with bioinsecticides. These longevities were found to be, significantly, reduced due to treatments, compared to those recorded from the control moths (6.2 & 6.2, 6.2 & 6.6 and 6.6 & 6.6 days for male and female from treatment by Dipel 2X, Dipel EC and Ecotech opposed to 8.8 & 9.2 for male and female from control.

## 1.2.g. Effects due to S. littoralis 6th instar larval treatments:

As shown in Table (15), it is clear that feeding of the sixth instar larvae of S. *littoralis* on castor-bean leaves treated with the LC<sub>50</sub>'s of Dipel 2X, Dipel EC and Ecotech caused 56, 54 and 53% mortalities among the treated larvae, respectively.

#### Larval period:

The surviving *S. littoralis* larvae after  $6^{th}$  instar treatments by bioinsecticides showed significant, prolongations in the larval period than control. The longest period of sixth instar  $(7.7 \pm 0.5 \text{ days})$  occurred due to Dipel 2X treatment, followed by  $6.8 \pm 0.4$  and  $6.4 \pm 0.4$  days from Dipel

Table (14): Effect of S. littoralis 5th larval instar treatment by B. thuringiensis preparations on fecundity and longevity of the resultant adults (Results from 3 pairs).

Treatment (I.U./mg).	Oviposition period (days)	Averag	Average no. of	%	Adult longevity (days)	vity (days) E.
	± <b>3.D.</b>	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	hatching	male	female
Dipel 2X	5.9 ± 0.1 b	368.0 ± 10.2 b	280.0 ± 7.2 °	76.09	6.2 + 0.2 b	62+0016
$(4.1440 \times 10^3)$	(5-6)	(350-390)	(250-300)		(6-7)	(6-7)
Dipel EC	$6.0 \pm 0.3$ b	380.0°± 7.2 b	$300.0 \pm 8.5$ bc	78.95	62+02b	66+016
$(4.6912 \times 10^3)$	(5-7)	(360-400)	(290-310)		(6-7)	(5-7)
Ecotech	6.2 ± 0.4 b	$390.0 \pm 7.2^{\ b}$	320.0 ± 4.5 b	82.05	66+01b	9 c 0 + 9 9
$(4.6944 \times 10^3)$	(5-7)	(360-420)	(300-340)		(5-7)	(6-8)
Control	8.7 ± 0.4 a	530.0 ± 13.6 a	483.0 ± 9.0 °	91.13	8.8 ± 0.2 a	9.2 ± 0.1 °
	(8-10)	(518-540)	(470 - 495)		(8-9)	(9-10)
F-(0.05)	16.02*	100.57*	215.07*		13.42*	35.26*
L.J.D. (0.05)	1.15	26.08	21.84		1.11	0.80

EC and Ecotech treatments, respectively, opposed to  $4.4 \pm 0.5$  days for the untreated larvae (Table, 15).

## Prepupal & Pupal mortalities & pupal and adults' malformations:

The data concerning mortality and malformation percentages amongst pupae and adult stages resulted from *S. littoralis* 6<sup>th</sup> instar larval treatments by the LC<sub>50</sub>'s of 3 bioinsecticides are recorded in Table (15). The mortality percentages among prepupae ranged from 11.32% by Ecotech treatment to 20.45% from Dipel 2X treatment, while those of pupae averaged 14.28% from Dipel 2X treatment to 15.91 % from Dipel EC treatment. While, the highest percentages of malformed pupae and adults were estimated as 13.3% and 19.23% from Dipel 2X treatment, and the lowest percentages ( 8 and 16.67%) from Ecotech treatment (Table, 15).

#### Pupal period:

As occurred with the previous (5<sup>th</sup>) larval instar, analysis of variance in pupal period between treated and untreated larvae were insignificantly different, although slight prolongations in this period due to Dipel 2X, Dipel EC and Ecotech treatments were detected (Table, 15).

# Fecundity and fertility of emerged moths:

Data presented in Table (16) indicate that feeding of the S. *littoralis*  $6^{th}$  instar larvae on castor-bean leaves treated with the LC<sub>50</sub>'s of the three bioinsecticides under study caused significant reductions in the fecundity and fertility of the subsequent adults. The oviposition period was found to be , significantly , reduced from 8.7 days for the control female to 6.1, 6.3 and 7.1 days due to Dipel EC , Dipel 2X and Ecotech

treatments, respectively. Also, the number of eggs productivity averaged  $400 \pm 7.2$ ,  $410 \pm 8.5$  and  $440 \pm 6.4$  eggs /female from the three bioinsecticidal treatments, respectively, being , significantly, fewer than that recorded from control ( $540.6 \pm 7.3$  eggs/female). The hatchability percentage was, also, found to be reduced from 89.53% among eggs deposited by control females to 80, 80.49 and 84.09% from eggs deposited by females resulted after  $6^{th}$  instar larval treatments by Dipel 2X, Dipel EC and Ecotech, respectively (Table, 16).

### Adults' longevity:

Table (16) shows the longevities of *S. littoralis* male and female adults treated in the 6<sup>th</sup> instar larva with three bioinsecticides. These longevities averaged 8.6 and 9.2 days for control male and female, respectively and were found to be, significantly, shortened to 6.2 & 6.4, 5.8 & 6.4 and 7.2 & 7.4 days for males & females resulted from larval treatment by Dipel 2X, Dipel EC and Ecotech at the LC<sub>50</sub> levels (Table, 16).

In agreement with the present results concerning the biocidal larval treatment on the survivors of S. *littoralis*,  $Salama\ et\ al\ (1981)$  reported that the  $2^{nd}$  instar larval treatment by Thuricide caused retardation in the larval development. Also,  $Gadallah\ et\ al\ (1990)$  found that S. *littoralis*  $4^{th}$  instar larval treatment by Dipel caused increase in the larval and pupal durations as latent effect. The former author found that Thuricide treatments to  $2^{nd}$  instar S. *littoralis* larvae caused significant reduction in pupal weight and appearance of deformities in subsequent pupa and moths' populations in addition to reduction in eggs productivity which reached 420 and 512 eggs / female due to rearing on diets containing 400 and 100 IU toxin / ml., respectively opposed to 798 eggs / a control

Table (16): Effect of S. littoralis 6th larval instar treatment by B. thuringiensis preparations on fecundity and longevity of the resultant adults (Results from 3 pairs).

(I.U./mg).	Oviposition period (days)	Averag	Average no. of		Adult longevity (days)	≀ity (days) €.
	1-	deposited eggs/ female ± S.E.	hatched larvae ± S.E.	% hatching	male	female
Dipel 2X	6.3 ± 0.4 b	400.0 ± 7.2 °	320.0 ± 3.2 °	80 00	40+04b	
$(5.9392 \times 10^3)$	(6-8)	(380-420)	(310-330)		0.2 TO +	6.4 ± 0.2°
Dipel EC	6.1 ± 0.2 b	410.0 + 8.5°	3300+720	00 40	(3-1)	(5-7)
$(8.4768 \times 10^3)$	(5-7)	(380-430)	(210 250)	00.47	5.8 ± 0.5	$6.4 \pm 0.3$ b
Ecotech	71+066	4400 - 0 - 6	(000 010)		(4-7)	(4-7)
(7 5711 163	7.1 - 0.0	440.0 ± 6.4 °	370.0 ± 9.1 °	84.09	$7.2 \pm 0.1$ ab	7.4 + 0.1 b
(7.5/44 x 10°)	(6-8)	(420-460)	(350-390)		(7-8)	(8.7)
Control	8.7 ± 0.4 a	540.6 ± 7.3 *	484.0 ± 5.2 °	89.53	86+02	02+02
	(8-10)	(520 –560)	(471-496)	35 4 4	(7 O)	7.2 ± 0.2
F. (0.05)	12.9*	))) 0)*			(1-9)	(8-10)
LSD	113	220.83"	45.69*		6.71*	11.47*
(0.03)	CI'I	22.91	38.33		1.66	1.34

female . Also , *Hafez ( 1993 )* found that *S. littoralis*  $1^{st}$  instar larval treatment by Thuricid ( Hb ) at the rate of  $30 \times 106$  IU caused 15 % mortality on the first day after treatment , and the resultant female moths deposited an average of 407 eggs / female opposed to 704 eggs / a normal female moth .

## 2. Efficacy of Dipel 2X on S. littoralis in the field:

Dipel 2X was applied, in the field, on August,  $4^{th}$  1998, at the recommended rate (200 gm./400 litres/feddan) on cotton plants that were artificially infested by the cotton leafworm egg – masses (at the rate of 4 egg masses / plot; *i. e.*, an egg mass / about 10 plants). Spraying took place just after distribution of the eggs in the field.

Random samples, of 75 cotton leaves each / plot were collected 7, 10 and 15 days after spraying and transported to the laboratory where each leaf was thoroughly examined for any sign of larval feeding. The rate of infestation was calculated according to *Kasoper's* formula (1965). Data are tabulated in Table (17).

It is clear from the mentioned table that spraying of Dipel 2X caused reductions in the rate of infestation by the cotton leafworm than control . The reduction percentages were 51.01, 56.31 and 56.56 % after 7, 10 and 15 days, of treatment, respectively. The overall mean of infestation was 2.93 in the control treatment which was found to be reduced due to bioinsecticidal treatment to 1.31 indicating an average reduction in the whole mean percentage of infestation by 55.29 %. These reduction percentages in the rates of infestation by *S. littoralis* is normally because of the decrease in larval population due to the effective biocontrolling role of *B. t. kurstaki* ( the main component of Dipel 2X).

In similar studies, *Salama* and *Zaki (1984)* studied the effect of Dipel ( *B. t. kurstaki* ) on *S. littoralis*. They found that using the bioinsecticide at 500 g/feddan on cotton, reduced the larval population but the egg masses of this pest were not affected by the treatment. While, *El-Husseini (1984)* applied Bactospeine ( *B. thuringiensis* var. *kurstaki* ) containing 16000 IU/mg, at a rate of 1 kg/feddan, in

Table (17): Efficacy of field application of Dipel 2X on the rates of infestation by S. littoralis larvae infesting cotton

Period after	Infestati	Infestation rate	% redaction due	Means	ans
treatment	control	treated	to treatment	C°	% R. H.
7 days	1.98	0.97	51.01	31.51	57.95
10 days	2.93	1.28	56.31	31.35	57.79
15 days	3.89	1.69	56.56	30.88	58.78
Mean	2.93	1.31	55.29		

fields of clover . This treatments gave only 5 % control of  $\emph{S. littoralis}$  larvae and pupae

Also *Shalaby et al (1993)* studied the effect of Delfin (a selective bacterial insecticide containing 53 X 10 <sup>6</sup> S. U. of *B. t.* var. *kurstaki* / gr of product), applied at a rate 0.525 Kg / feddan. The overall average of damage caused by *S. littoralis* was reduced by 18.86 % due to the application of the bioinsecticide. *Abdel – Halim (1997 b)* studies the efficacy of Dipel 2X (32,000 IU *B. t. kurstaki* / mg), when used at a rate of 200 gm / feddan, against *S. littoralis* larvae infesting cotton and Egyptian clover. The author found that treatment caused initial mortality of 79.8 % after 7 days from treatment. *El-Husseini et al (1997)* reported 42.8 % mortality among *S. littoralis* larvae infesting clover after 10 days of spraying Bactospeine EC (*B. t. kurstaki*; 13000 IU / mg) at a rate of 0.5 L / feddan.

# 3. Evaluation of some materials as UV photoprotectants for increasing the efficacy of *B. thuringiensis*:

#### 3.1. Laboratory studies:

Three different materials; shellac, melanin and neste – coffee were assayed for their capability to absorb U V rays and subsequently increasing the efficacy of *B. thuringiensis kurstaki* against *S. littoralis* larvae. The absorption spectra of the three mentioned materials were previously assayed by *Ragaei* (1998), *Shams El-Din* (1998) and *Ragaei* (unpublished work), respectively. According to their results, the absorption spectra of each of these materials at 1% concentration, when exposed to different wave lengthes from 254 to 560 nm. are presented in Table (18).

From data tabulated in Table (19), it is clear that feeding the 1<sup>st</sup> instar larvae of **S. littoralis** for 48 hours on castor – bean leaf – discs treated with **B. t. kurstaki** (Dipel 2X at 0.5 % concentration) alone or that mixed with flour + yeast , and that mixed with flour + yeast + 1% of either of the three photoprotectant materials resulted mortality percentages ranged from 65-67 %. By exposure of Dipel 2X and the tested mixtures to UV for 4 hours , the efficacy of **B. t. kurstaki** was found to be reduced by 46.15 %, as the mortality percentage decreased to 35 %. In case of mixing the bioinsecticide with flour and yeast , the efficacy was decreased by 40.43 % (47 % mortality after 5 days of starting treatment).

By adding either of the three photoprotectant materials (shellac, melanin or neste – coffee) at 1 %, the reduction in efficacy due to exposure to UV for 4 hours decreased greatly, being only 3, 4.48 and 4.62 %, respectively (64, 64 and 62 % mortality, respectively; Table,

Table (18): Absorption spectra of shellac, melanin and neste-coffee at 1% concentration to U.V. at different wave lengths.

Wavelengths (nm)							
Materials	254	290	320	340	365	364	Remarks
Shellac	3.67	3.64	3.64	3.64	3.25	1.01	(after Ragaei, 1998)
Melanin	3.67	3.67	3.64	3.64	3.70	3.74	(after Shams El-Din, 1998)
Neste - coffee	3.67	3.64	3.64	3.64	3.77	2.42	(after Ragaei unpublished)

19). These results confirmed that shellac, melanin and neste – coffee are excellent U V protectant materials.

By elongation of the exposure period to U V up to a maximum of 16 hours, data in Table (19) indicated, generally, a negative relationship between the exposure period and the efficacy of B. t. kurstaki; i.e., the efficacy (calculated as larval mortality % after 5 days of treatment) decreased by the increase of the exposure period to UV But, the rate of decrease in efficacy was, greatly, reduced by using either of the three assayed photoprotectants. By exposure to UV for 16 hours, the efficacy of B. t. kurstaki decreased by 69.2 % as the larval mortality percentage decreased to only 20 % opposed to 65 % by using unexposed Dipel 2X; i.e. B. t. kurstaki lost more than 2/3 of its activity due to exposure to UV for 16 hours. By adding flour and yeast to the bioinsecticide, the percentage reduction in efficacy reached 54.5 % (30 / 66). While, on the contrary, in case of adding shellac, melanin or neste – coffee to envelope the spores of **B. t. kurstaki**, these materials were able to protect the spores from the direct injury of long exposure (16 hours) to U V, and the reductions in the efficacy of the product reached 7.6 % (61 /66), 7.5% (62 / 67) and 12.3 % (57 / 65) ,respectively (Table, 19).

The remaining activity of the tested bioinsecticide (Dipel 2 X) with and without mixing by the photoprotectants after exposure to UV light could be expressed as original activity remainder (O A R); the increase in this value indicates more activity of the compound. That value was easy to be calculated by considering the efficacy of  $\textbf{\textit{B.t.}}$  kurstaki alone (before exposure to U. V.) as 100 %, and then efficacy of the different assayed materials could be, subsequently, calculated as a rate to the 100 % of the unexposed bioinsecticide.

Table (19): Larval mortality percentages among 1st instar S. littoralis larvae fed on castor - bean leaves treated with B. presence of photoprotectants. (Data recorded after 5 days from treatment of 100 larvae / treatment). thuringiensis subsp. kurstaki (Dipel at 0.5 % concentration ) irradiated with U. V. rays in absence and the

	% of larva	l mortality afte	% of larval mortality after exposure of products to U.V. (in hours)	oducts to U.V. (	(in hours)
Treatment	0	4	<b>∞</b>	12	16
B. t. alone	65:00	35.00	30.00	25.00	20.00
Flour + yeast + B. t.	66.00	47.00	41.00	35.00	30.00
Flour + yeast +B. t. + 1% shellac	66.00	64.00	63.00	63.00	61.00
Flour + yeast + B. t. + 1% melanin	67.00	64.00	63.00	58.00	62.00
Flour + Yeast + B. t.+1% neste-coffee	65.00	62.00	60.00	59.00	57.00

As seen in Table (20), by expose of Dipel 2X alone or mixed with the mentioned materials to U V for 16 hours, the original activity remainder (OAR) of **B.** t. kurstaki alone was greatly reduced to 30.77, being less by more than 1/3 of that of the unexposed **B.t.** kurstaki. Thus confirming the detrimental effect of U.V. light on the spores of **B.** thuringiensis. In this respect, Ignoffo and Batzer (1971) and Pozsgay et al (1987) reported that irradiation of U V light at wave — lengths from 250 nm had a detrimental effect on the viability and toxicity of **B.** thuringiensis.

The obtained O A R from using Dipel 2X alone after exposure to (30.77), increased to 46.15 when flour and yeast were added to the bioinsecticidal product, showing a limited useful effect for protection from U V light which appeared as some increase in the remaining activity of the product. While, by adding either of the three tested materials ( shellac , melanin or neste - coffee ) at 1 % concentration to the bioinsecticide (Dipel 2X + flour + yeast), high increases in the O A R occurred to reach 93.85, 95.38 and 87.69, respectively (Table, 20). These high OAR's indicate that B. t. kurstaki spores kept most of their activity inspite of exposure to the U. V. light for 16 hours. This high resistance to U. V. is, undoubtedly, attributed to the role of the photoprotecting materials in encapsulating the B. thuringiensis spores and absorping the U. V. light before reaching the spores . From data in Table (20), melanin and shellac could be, fairly, considered as excellent photoprotectant materials as they kept 95.38 and 93.85 % of the original activity of B. t. kurstaki after 16 hours exposure to U.V., while, neste-coffee at 1 % concentration proved as a very good photoprotectant as the O A R after exposure was lower, being 87.69. In agreement with these results, Liu

Table (20): Original activity remaining (OAR) of B. thuringiensis subsp. kurstaki (Dipel 2X) with & without photoprotectant after exposure to U. V. irradiation for 16 hours against S. littoralis larvae.

Treatment % mor	% mortality of <i>S. littoralis</i> larvae	Original activity remaining (O A R)
B. t. alone (control ) (without exposure )	65.00	100.00
After exposure		
B. t. alone	20.00	30.77
Flour + yeast + B. t.	30.00	46.15
Flour + yeast + B. t. + 1 % shellac	61.00	93.85
Flour + yeast + B. t. + 1% melanin	62.00	95.38
Flour + yeast + B. t. + 1% neste - coffee	57.00	87.69

et al (1993) indicated that melanin showed an excellent photoprotective potency for **B.** thuringiensis var. israeliensis since it gave broader protection than any other sunscreen material, it acts as a neutral density filter, scatters impigning radiation, absorbs energy in the UV, acts as a trap for unstable electrons, and prevents the formation of unstable free radicals. The authors reported that melanin has its own way to protect the bacteruim, **B.** thuringiensis against U.V.

In accordance with the presented results, *Possagy et al (1987)* showed that exposure of *B. thuringiensis* to U V irradiation caused destruction of 80 % of tryptophan and 20 % of histidine residues and suggested a photosensitization mechanism by which an exogenous chromophore absorps light and transfers energy to an  $O_2$  molecule. *Ephraim Cohen et al (1991)* reported that irradiation of *B. thuringiensis* var. *kurstaci* HDI at 300-350 nm for up to 12 h. resulted in a rapid loss in toxicity to larvae of *Heliothis armigera*. The author attributed the detrimental effect of U V irradiation to inactivation of the  $\delta$  endotoxin. The same authors speculated also that the destruction of tryptophan residues may result in profound changes in the three – dimentional configuration of the toxic protein and consequently the loss of its biological activity.

In a study similar to the present, *Ragaei* (1998) assayed the efficacy of *B. thuringiensis* var. *entomocidus*, after being exposed to U V for 16 hours, on neonate larvae of *S. littoraris*. The author found that U V irradiation caused 68.75 % reduction in the efficacy of the bacterium ( percentage mortality was reduced from 96 to 30 %). When the author used the same variety of bacreria + shellac 1 %, the mortality % among larvae was reduced by only 11.45 % (85 % mortality); *i. e.*, the O A R

due to shellac treatment reached 88.5 %. This value increased to 93 % by increasing the shellac concentration to 2 %. The effect of melanin at % concentration, as a photoprotectant from U V irradiation for increasing the efficacy of B. thuringiensis var. entomocidus, was studied by Shams El - Din (1998) who found that adding of melanin 1  $\%\,$  to  $\,$  the bacterium and exposure to U V for 16 hours led to 97 O A R ; i. e. 97 % of the efficacy of B. thuringiensis was saved, while the O A R in case of exposure of **B.t. kurstaki** alone to U V for the same period was only 31; i.e., the bacterium lost 69 % of its efficacy due to irradiation. While, the useful role of neste – coffee as a photoprotectant the detrimental effect of U V irradiation on B. t. entomocidus was studied by Ragaei (unpublished work). The author found that exposure of **B.** t. entomocidus alone to U V for 16 hours inhibited the activity of entomopathogenic bacteria and the mortality % among treated S. littoralis neonate larvae was reduced from 82 % (by using unexposed bacteria ) to only 30 % when the bacteria were exposed to U V before treatment (OAR 36.6). While, by adding neste – coffee to the bacteria and exposure to U V for the same period, the O A R was found to be increased to 93, as percentage mortality among treated larvae was 76 %.

#### 3.2. Field studies :

The forementioned studies confirmed the detrimental effect of U V irradiation on  $\boldsymbol{B}$  .  $\boldsymbol{t.}$  kurstaki and the possibility of avoidance of this effect by using any of the photoprotectants (shellac, melanin or neste – coffee ) to absorb the U V rays and, subsequently, avoid the bad effect of U. V. on  $\boldsymbol{B.}$  thuringiensis . Accordingly, it was necessary to study the possibility of using the photoprotectant materials under study in the

field to find out their role as protectants for the entomopathogenic bacteria under natural conditions.

Dipel 2X (*B. t. kurstaki*) alone and that mixed with flour + yeast or with flour + yeast + either of three photoprotectants were sprayed on cotton plants. Treated cotton leaves were picked just after spraying and after 24, 48, 72 and 120 hours, brought to the laboratory and offered to *S. littoralis* first larvae in ½ kg. glass containers for 48 hours feeding, then the surviving larvae were allowed to continue feeding on fresh untreated cotton leaves until pupation. Mortalities were measured after 5 days from treatment, whereas the resultant moths from each treatment were paired to study the fecundity of these adults.

Data presented in Table (21) demonstrates the efficacy of Dipel 2X ( B. thuringiensis kurstaki ) estimated as percentages of 1st instar S. littoralis larval mortalities after different periods of field spraying on cotton plants. It is clear from the mentioned table, that at 0 time (just after bioinsecticidal spraying ), all the applied materials either the bioinsecticide alone or mixed with different materials caused 64 – 66 % mortalities among the treated larvae indicating, nearly, equal efficacy of the applied materials. This may due to at that time, the bacteria were not yet exposed to the environmental conditions . 24 hours later, the efficacy of Dipel 2X was found to be decreased by 43.75 % as it caused only 36 % mortality . Adding flour + yeast to Dipel 2X caused little improvement in the activity of **B. t. kurstaki** as the percentage of 1st instar S. littoralis larval mortality decreased from 65 % at zero time to 47 % (27.7 % reduction) after 24 hours of treatment. While, by adding shellac, melanin or neste - coffee at 1 % to the bioinsecticide + flour + yeast resulted in high protection of the bacterium activity against S.

Table (21): Efficacy of Dipel 2X (B. t. kurstaki) on S. littoralis neonate larvae after different period from field treatment . ( Data from treatment of 100 larvae ) .

	% то	tality of s	% mortality of <i>S. littoralis</i> larvae	120	after U.V. exposure	posure		
Treatment			period afte	period after treatment	•		Mean	an
	0	24 h.	48 h.	72 h.	96 h.	120 h.	C°	% R. H.
B. t. alone	64.00	36.00	25.00	20.00	16.00	10.00	32.09	57.67
Flour + yeast + B. t.	65.00	47.00	35.00	30.00	23.00	20.00	32.54	54.71
Flour + yeast + B. t. + 1% shellac	66.00	60.00	56.00	52.00	50.00	45.00	32.22	58.21
Flour + yeast + B. t. + 1% melanin	66.00	59.00	54.00	50.00	46.00	42.00	30.94	63.04
Flour + yeast + B. t + neste - coffee	64.00	56.00	50.00	45.00	40.00	35.00	32.22	59.71

littoralis, as the respective mortality percentages were 66, 66 and 64% just after treatment and decreased after 24 hours to 60, 59 and 56% indicating reduction in the activity of  $\textbf{\textit{B.t. kurstaki}}$  by only 9.1, 10.6 and 12.5% respectively. Thus confirmed the protecting action of shellac, melanin and neste – coffee, especially against the U.V. irradiation emitted by the sun .

As shown in Table (21), the efficacy of B. t. kurstaki, alone and that mixed with the mentioned additives, against S. littoralis neonate larvae ( as percentages of larval mortality ) was estimated daily in the laboratory by using leaves picked from cotton plants throughout the successive 5 days after spraying. It is clear from this table that the potentiality of bacteria decreased sharply and successively when the bioinsecticide (Dipel 2X) was used alone, as the percentage mortality reached only 10 % after 5 days exposure in the field, indicating 84.4 % reduction in the bacterium activity; and accordingly the OAR value was only 15.6 ( Table , 22 ). In case of adding flour + yeast to Dipel 2X, then spraying on cotton plants, leaving the spray under field conditions for 5 days and using cotton leaves to feed S. littoralis neonate larvae, the mortality % was found to be reduced from 65 % (by using leaves at zero time ) to 20 % (69.2 % reduction in B. t. kurstaki activity ) , indicating an OAR of 31.25 %. By adding shellac , melanin or neste - coffee at 1 % to the previous mixture and estimating the activity in the laboratory after exposure for 4 and 5 days, the obtained percentages of S. littoralis larval mortality reached 50 & 45 %, 46 & 42 % and 40 & 35 % , respectively indicating reduction in the bacterium activity by 24.2 & 31.8, 30.3 & 36.4 and 37.5 & 45.3 %, respectively ( Table ,21) . The calculated O A R's of the 3 previous mixtures after 4 & 5 days exposure to field conditions were 78.13 & 70.31, 71.88 &

Table (22): Original activity remaining (OAR) of Dipel 2X (B. t. kurstaki) with and without photoprotectants against S. littoralis after exposure to field conditions for 4 and 5 days.

Tractment	% mortality of S. littoralis larvae	S. littoralis larvae	Original activity remaining (OAR)	maining (OAR)
TICALIICIIC	after 4 days	after 5 days	after 4 days	after 5 days
B. t. alone (control )( without exposure )	64.00	64.00	100.00	100.00
After exposure	Đ.			
B. t. alone	16.00	10.00	25.00	15.63
Flour + yeast + B. t.	23.00	20.00	35.94	31.25
Flour + yeast + $B$ . $t$ + 1 % shellac	50.00	45.00	78.13	70.31
Flour + yeast + $B \cdot t + 1\%$ melanin	46.00	42.00	71.88	65.63
Flour + yeast + B. t. + neste - coffee	40.00	35.00	62.50	54.69

65.63 and 62.50 & 54.69, respectively (Table, 22). These data proved that shellac may be, fairy, considered as the best photoprotectant as it led to the lowest reduction in **B. thuringiensis** potentiality *i. e.*, highest **S. littoralis** mortality percentages and highest Original activity remainders (OAR's) after exposure to field conditions, followed by melanin. While neste—coffee gave rates of protection of the bacterium against natural field conditions lower than those estimated in cases of shellac and melanin. But, in all cases the obtained field results confirmed those of laboratory as the three materials proved as good photoprotectants of **B. thuringiensis** and accordingly either of these materials may be recommended as to be mixed with the **B. thuringiensis** commercial products for protection of this entomopathogenic bacteria, after spraying in the field, especially against the detrimental effect of the sun U.V.

#### 3.2.a. Effect on larval, prepupal and pupal mortalities:

As shown in Table (23), feeding of *S. littoralis* 1<sup>st</sup> instar larvae on cotton leaves just after spraying the cotton plants in the field (at zero time) by Dipel 2X alone at the recommended concentration (0.5%), mixed with flour + yeast or mixed with the same materials + 1% shellac, melanin or neste – coffee led to 65-67, 25.71-30.55 and 11.54-20% mortalities during the larval, prepupal and pupal stages, respectively, opposed to 0, 2 and 1.02% mortalities among the same stages when larvae were fed after hatching on untreated cotton leaves.

When the applied materials ( **B.** t. kurstaki preparation and the mixtures ) were left on cotton plants in the field for 24 hours after which treated cotton leaves were offered to **S.** littoralis 1<sup>st</sup> instar larvae, the

Table (23): Larval & pupal periods and mortality percentages, and pupal & adults' malformations due to of S. littoralis 1st instar larval feeding on treated cotton leaves with Dipel 2X (B. t. kurstaki), alone and mixed with photoprotectants , at zero time . ( Rearing at 27  $C^{\circ}$  and 72 % R.H. ) .

Treatment	% n	% mortality among	ong	Durations	Durations (in days)	% malformations	rmations
	larvae	prepupae	pupae	larvae	pupae	pupae	adults
R + alone	65 00	25 71	11 54	$20.0 \pm 0.2^{\ b}$	$12.6 \pm 0.2^{b}$	19 23	13 04
	00.00	FO.1.1		(18.3 - 20.3)	18.3 – 20.3) (11.0 – 14.0)	17:20	10.0
Flour + Woost + R	66.00	78 57	20 00	20.6 ± 0.5 b	$12.8 \pm 0.2^{\ b}$	15 00	17 65
FIGUI - yeast - D	00.00	10:01	20:00	(18.0 – 24.0)	(12.0 – 14.0)	10.00	17.05
Flour + veast + R / + 1% shellar	66 00	30 55	20 00	$20.7 \pm 0.4^{\ b}$	$13.0 \pm 0.2^{\ b}$	24 00	25 00
FIGURE 1 years 1 20 to 1 1/0 SHORAN	00.00	00:00	10.00	(18.3 – 23)	(12.0 – 14.0)	17.00	10.00
Flour + vesst + $RI + 1\%$ melanin	67 00	29 41	16 67	20.1 ± 0.4 b	$12.8 \pm 0.1^{\ b}$	20.83	20 00
rioul - Jeast - 20 to - 1 /o meanin	0.00	t)		(18.5-23.0)	(12.0 - 13.0)	10.00	10.00
Flour + worst + R / + 10/ nost - coffee	65 00	25 71	11 54	$20.0 \pm 0.2^{\ b}$	$12.6 \pm 0.2^{\ b}$	19 23	13.00
FROM 1 years 1 D. 6 1 1 /0 Heat - collect	05.00	20.71	11.54	(18.0 –23.3)	(12.0 – 13.0)	17.25	15.00
Control	0 00	2 00	1 00	15.4 ± 0.1 a	8.0 ± 0.1 a	0 00	0 00
Connec	0.00	200.0	1.02	(15.0 – 16.0)	(7.0 – 9.0)	0.00	0.00
F. (0.05)				8.13*	32.67*		-
L.S.D. <sub>(0.05)</sub>				1.98	0.92		

mortality percentages were found to be reduced to 36, 20.41 and 12.82 % among larvae, prepupae and pupae, respectively in case of using Dipel 2X alone (Table, 24); i. e., 44.6, and 20.6 % reductions in the % mortality percentages among larvae and prepupae than those recorded the zero time treatment, while no decrease occurred in the percentage of pupal mortality. In case of Dipel 2X + flour + yeast, the mortality percentages reached 47, 22.64 and 12.19 %, respectively showing 28.8, 20.8 and 39.1% reductions than those recorded for the zero time treatment. These results confirmed the detrimental effect of **B.** thuringiensis exposure to environmental conditions (especially sun light) on the efficacy of the product. When shellac, melanin or neste – coffee was added at 1 % concentration to the mixture of Dipel 2X + flour + yeast , the reduction percentages in larval , prepupal and pupal mortalities were found not greatly affected when the mixtures were exposed for 24 hours to natural field condition after treatments. These mortality percentages were 60, 25 and 16.67 % for shellac, 59, 21.95 and 16.13 % for melanin and 56, 22.73 and 17.65 % for neste - coffee among larvae, prepupae and pupae, respectively. Comparing these percentages with those obtained from the zero time treatment, it could be deduced that the reduction in mortality percentages were 9.0, 18.2 & 16.7 % in case of shellac and 11.9, 25.4 & 3.2 % for melanin for the three mentioned stages, respectively. While in case of neste – coffee the mortality % decreased by 13.8 and 11.6 % for larvae and prepae, but that of pupae increased by 52.9 % (Table, 23 & 24). Thus indicating, generally, less reduction in the mortality %, due to exposure to sun light, by adding either of three photoprotectants to the bioinsecticide, than using the product alone.

From results tabulated in Table (25), more detrimental effect on the efficacy of Dipel 2X occurred by elongation of the exposure period

Table (24): Larval & pupal and adults' emergence from durations, mortalities and malformations due to S. littoralis 1st instar larval feeding on field treated cotton leaves with B. t. kurstaki alone and that mixed with photoprotectants ( 24 h. after bioinsecticidal application).

Trootmont	% г	% mortality among	ong	Durations	Durations ( in days )	% malformations	rmations
	larvae	prepupae	pupae	larvae	pupae	pupae	adults
B , along	36 00	20 41	12 82	17.5 ±0.2 °	10.8 ± 0.2 °	5 13	0 37
D. I. Alville	00.00	17.03	12.02	(17.4 – 20.4)	(11.0 – 12.7)	9.13	
Flow + coost + R	47 00	22 64	12 10	$19.2 \pm 0.4^{\ b}$	12.0 ± 0.1 b	8 33	0 00
FIGUIT YEAST D.A.	77.00	10:44	12.17	(18.0 - 20.0)	(11.0 – 13.0)	0.55	2.02
Flour + wast + R / + 1% shellar	60 00	25 00	16 67	$19.9 \pm 0.2^{b}$	$12.3 \pm 0.3^{\ b}$	10 00	13 64
FIOUI - yeast - D. 6 1 /0 SHCHAC	00.00	10.00	10.07	(19.4-20.2)	(12.0 - 13.0)	10.00	10.01
Flour + worst + R + + 10/ molonin	00 00	21 95	16 13	$19.4 \pm 0.3^{\ b}$	$12.1 \pm 0.1$ b	6 45	12 50
Flour - yeast - D. t 1 /o IIIciaiiii	0.00	11.55	10:10	(17.5-20.8)	(11.0 – 12.8)	9.10	12:00
Flour + west + R + + 10/ neste - coffee	56 00	22 73	17 65	$19.4 \pm 0.4^{\ b}$	12.1± 0.2 <sup>b</sup>	5 88	11 54
FIOUR - yeast - D. E 1 /o neste - contec	0.00	2.1.2	17.00	(18.0 – 20.9)	(11.0 – 13.2)	0.00	11.07
Control	0 00	0 00	1 00	$15.2 \pm 0.1$ a	$7.8 \pm 0.1$ a	0 00	0 00
Connec	0.00	0.00	1.00	(15.0-16.0)	(7.0 - 9.0)		0.00
F. (0.05)				11.81*	22.90*		
L.S.D. <sub>(0.05)</sub>				1.06	0.94		

to natural field conditions to two days. Much lower mortality percentages were obtained among the *S. littoralis* larvae, prepupae and pupae treated in their 1<sup>st</sup> instar larvae (25, 16.67 & 10 % by using *B. t. kurstaki* alone and 35, 15.38 & 18.18 % for Dipel 2X + flour + yeast among individuals of the three stages, respectively). While, when shellac, melanin or neste – coffee were added at 1 % to the mixture of the bioinsecticide + flour + yeast and exposure for 48 hours to field conditions, much higher mortality percentages were recorded among the three stages, although the recorded percentages were somewhat lower than those in case of treatment at zero time (56, 22.72 & 14.7 % for shellac, 54, 20.83 & 13.16% for melanin and 50, 20 & 15 % for neste – coffee treatment, respectively; Table, 25).

When the exposure period of Dipel 2X and the mixture, to natural field conditions, was extended to 3 days after spraying, severe detrimental effect on B. t. kurstaki occurred which appeared as acute decrease in the recorded mortality percentages (increased survivors) among the immature stage individuals after feeding the first instar S. littoralis larvae on cotton leaves treated with Dipel 2X or with Dipel 2X + flour + yeast for 48 hours followed by feeding on untreated leaves ( only 20, 12.82 and 7.35 % in the former treatment and 30, 14.28 and 11.67 % in the latter one among the three stages, respectively; Table, 26 ) . Comparing results of these two treatments with those obtained by treatment at zero time, it could be deduced that exposure of the bioinsecticide alone or mixed with flour and yeast for 3 days to field conditions resulted in reductions in the recorded mortalities than those recorded from treatment at zero time by 69.2, 50.1 & 36.3 % in former treatment and 54.5, 50 & 41.7 % in the latter one among larvae, prepupal and pupae, respectively. By adding either of the three

Table (25): Larval & pupal periods and mortality %, and pupal and adults' malformations due to S. littoralis larval bioinsecticidal spraying). feeding on field treated cotton leaves with Dipel 2X alone and mixed with photoprotectants (after 48 h. of

Tractment	١ %	% mortality among	ong	Durations (in days)	(in days)	% malformations	rmations
	larvae	prepupae	pupae	larvae	pupae	pupae	adults
- 1	25 00	16 67	10 00	$17.5 \pm 0.2$ °	9.8 ± 0.2 °	4 00	4 65
D. L. AIOHE	23.00	10.07	10.00	(16.4 - 18.7)	(10.0 - 11.0)	1.00	1.00
Florest constant	35 00	15 38	18 18	$18.0 \pm 0.4$ bc	10.2± 0.1 °	4 44	80 9
FIGUR T yeast T D		10.00	10.10	(15.0 – 20.0)	(8.0 – 10.7)	1.77	0.70
Flower transact t B + 10% shallon	26.00	22 72	14 70	$19.7 \pm 0.2^{\ b}$	$11.3 \pm 0.3^{\ b}$	8 87	11 54
FIGURE T YEAST T D. 6. T 1 70 SHEHAC	00.00	11.11	77.70	(18.5-21.0)	(11.0 - 13.2)	0.01	I
Flour t wood t B + 10/ molonin	54.00	20 83	13 16	18.4 ±0.4 bc	10.1 ± 0.1 °	5 26	9 68
FIOUF T yeast T D. L. T 1 /0 III CIAIIII	04.00	20.03	15.10	(17.7-20.0)	(9.0 – 11.0)	0.4.0	
Flour t spect t B + 10/ most coffee	\$0.00	20 00	15.00	$18.5 \pm 0.4^{\text{ bc}}$	10.2 ± 0.2 °	5 00	9 38
FIGURE T yearst T. L. T. 1 /0 HEST - COHECE	00.00	20.00	10.00	(15.9–20.8)	(9.0 – 11.6)	0.00	7.50
Control	1 00	202	2 02	15.2 ± 0.1 a	$7.8 \pm 0.1^{a}$	0 00	0.00
Control	1.00	1.01	1.01	(15.0-16.0)	(7.0 – 8.0)	0.00	
F. (0.05)				4.84*	8.01*		
L.S.D. <sub>(0.05)</sub>				1.84	1.09		

photoprotectantes under study to the mixture, much higher mortality percentages were obtained (lower numbers of survivors; *i. e.*, more efficacy than in case of the absence of these materials). After 3 days exposure to field conditions, the recorded mortality percentages among larvae, prepupae and pupae were 52, 20.41 & 12.82 % in case of shellac, 50, 18.18 & 11.11 % for melanin and 45, 17.54 and 12.76 %, respectively in case of neste – coffee (Table, 26). It is clear that the percentages of reduction in mortality percentages than zero time treatment were 21.2, 33.2 & 35.9 % for shellac, 25.3, 38.2 & 33.4 % for melanin and 30.8, 31.8 & 10.6 % in case of neste – coffee among the three stages, respectively.

#### 3.2.b. Effect on larval and pupal durations :

S. littoralis 1<sup>st</sup> instar larvae were fed on treated cotton leaves, from different treatments for 48 hours after which the survivors were supplied with untreated leaves until pupation. The larval and pupal periods were estimated.

As previously indicated in the first part of this study, feeding the first instar S. *littoralis* larvae on B. t. kurstaki treated leaves caused significant elongations in the larval and pupal periods of the survivors. This result was confirmed in the present experiment which showed that feeding the first instar larvae on cotton leaves treated with B. t. kurstaki alone or mixed with the additives under study, just after spraying in the field, caused significant prolongations in the larval and pupal periods than control (20-20.7 days for larvae and 12.6-13 days for pupae of treatments opposed to 15.4 and 8 days for larvae and pupae of control, respectively; Table, 23).

Table (26): Larval & pupal durations and mortality %, and pupal & adults' malformations due to S. littoralis 1st instar larval of application). feeding on field sprayed cotton leaves with Dipel 2X alone and mixed with photoprotectants (after 72 h.

Treatment	% п	% mortality among	ong	Durations (in days)	(in days)	% malformations	mations
	larvae	prepupae	pupae	larvae	pupae	pupae	adults
R / glone	20 00	12 82	7 35	$15.4 \pm 0.3^{a}$	8.0 ± 0.2 a	2 04	3 78
	10:00	12:02	ij	(14.0 - 17.0)	(7.0 – 9.0)	1.54	0.2.0
Flour + veget + R /	30 00	14 28	111 67	16.4 ± 0.5 ac	9.0 ± 0.2 bd	3 77	10.00
A TOWN . June . De	. 00	17.20	11.07	(15.0 - 18.0)	(8.0 - 10.0)	9.77	10.00
Flour + veast + $R$ / + 1% shellar	00 05	20 41	12 82	$19.2 \pm 0.2^{\ b}$	$10.2 \pm 0.1$ °	7 60	89.0
A TOWN JUNE 1 MARK A TO SHOULD	01:00	20.11	14.04	(18.0-21.0)	(10.0 - 11.0)	7.67	
Flour + vesst + $R$ $t$ + 1% melanin	50 00	18 18	=======================================	$18.0\pm0.2^{6}$	$10.0 \pm 0.1$ °	4 44	7 89
A TOTAL . JUNE . M. M. A. /O HILVERHILL	0.00	10.10		(16.0-19.0)	(9.0 – 10.0)	1	7.07
Flour + vesst + $R$ t + 1% nest - coffee	45 00	17 54	12 76	$17.0\pm0.5^{\ bc}$	$9.8\pm0.2^{\mathrm{cd}}$	4 25	7 60
A ACCORD - Joseph - AZO DO - A ZO MOSTE - COLLEGE	5.00	17.57	12:70	(15.0-21.0)	(9.0-11.0)	7.4.7	.00
Control	1 00	101	0 00	$15.0 \pm 0.1$ a	7.8 ± 0.2 a	000	0.00
Control	1.00	1.0.1	0.00	(14.0 - 16.0)	(6.0 – 9.0)	0.00	0.00
F. (0.05)				7.37 *	8.76 *		
L.S.D. <sub>(0.05)</sub>				1.57	0.95		

By lengthening the exposure period , of the sprayed materials in the field , to 24 hours before offering the treated cotton leaves to 1st instar S. *littoralis* larvae , the recorded larval and pupal periods of the survivors were found also , significantly , longer than that of the control treatment ( Table , 24 ) . But , in this case , the recorded periods from treatments were , generally , shorter than those recorded from the zero time treatments . Comparing the larval and pupal periods from different , treatments , data in Table (24) show that the periods recorded from treatment of Dipel 2X alone (  $17.5 \pm 0.2$  and  $10.8 \pm 0.2$  days for larval and pupal periods , respectively ) were , significantly , shorter than those recorded from either of the remaining 4 treatments of which the differences between them were insignificant ( 19.2 - 19.9 days for larvae and 12 - 12.3 days for pupae ; Table , 24 ) . Thus indicating that Dipel 2X alone was the most affected by field conditions ( especially sun rays ) even when exposed for only 24 hours compared to the other 4 mixtures .

More effect occurred when the bioinsecticide and the mixtures were left for 48 hours exposure to field conditions although all the recorded larval and pupal periods recorded from treatments ( 17.5-19.7 days for larvae and 9.8-11.3 days for pupae ) were significantly longer than those recorded from the control treatment ( 15.2 and 7.8 days , respectively ) ( Table , 25 ). Comparing between different treatments , the highest efficacy occurred when shellac was used as photoprotectant , as this treatment caused the longest larval and pupal periods (  $19.7\pm0.2$  and  $11.3\pm0.3$  days , respectively ) . While , on the other hand , Dipel 2X alone was the least effective ( most affected by field conditions ) as it caused the shortest periods (  $17.5\pm0.2$  and  $9.8\pm0.2$  days , respectively ; Table , 25 ) .

Prolongation of the exposure period of Dipel 2X and the mixtures to field conditions for three days after spraying caused another decrease in the effect of Dipel 2X and the 4 mixtures on the larval and pupal durations of S. littoralis ( Table , 26 ) . But , the highest efficacy of field conditions (mainly sun light ) occurred when the bioinsecticide was sprayed without any additives as the recorded periods were  $15.4 \pm 0.3$ and  $8 \pm 0.2$  days for larvae and pupae, respectively, which were insignificantly different that those recorded from the control treatment ( 15  $\pm$  0.1 and 7.8  $\pm$  0.2 days ; Table , 26 ) . As for the effect on Dipel 2X mixed with flour and yeast, this treatment caused insignificant prolongation (than control ) in the larval period (  $16.4 \pm 0.5$  days ) and significant prolongation in the pupal period ( $9 \pm 0.2$  days). While, adding either of the three photoprotectant materials caused decreased effect of exposure to field conditions, as all the estimated larval and pupal periods (  $19.2 \pm 0.2 \& 10.2 \pm 0.1$  days in case of shellac ,  $18 \pm 0.2$ &  $10 \pm 0.1$  days for melanin and  $17 \pm 0.5$  &  $9.8 \pm 0.2$  days by using neste - coffee, for larvae and pupae, respectively; Table, 26) were, significantly higher than those recorded from the untreated check.

The presented data concerning the effect of field spray of Dipel 2X alone or that mixed with different additives and exposure of this bioinsecticide to field conditions for different periods on the efficacy of **B. t. kurstaki** on the **S. littoralis** larval and pupal durations after feeding its 1<sup>st</sup> instar larvae on treated cotton leaves confirmed that previously demonstrated for the effect on immature stage mortalities. The well known effect of prolongation of larval and pupal durations due to **B. t. kurstaki** treatment was also detected in this experiment by treatment just after spraying. But, this effect decreased with the increase of exposure period, so that the biocidal effect, nearly, disappeared after 3 days of treatment (insignificant value than control by spraying Dipel 2X alone

or mixed with flour and yeast). While, by adding shellac, melanin or neste—coffee to the latter mixture kept, to great extent, the viability of **B. t. kurstaki** which appeared as significant prolongations in the larval and pupal durations even after 3 days exposure of the spray to field conditions. These results also confirmed the excellent effect of shellac, melanin and neste—coffee as photoprotectants for the mentioned biocontrolling pathogen. Insignificant differences were detected between the efficacies of the three materials in protecting **B. t. kurstaki** against U V emmited by sunlight.

# 3.2.c. Effect on % malformations among the obtained S. littoralis pupae and adults :

The resultant pupae and adults from different treatments were thoroughly inspected for counting the malformed individuals. Feeding of the 1st instar S. littoralis larvae on cotton leaves just after spraying Dipel 2X alone, the bioinsecticide + flour and yeast or that mixed with flour + yeast + either of shellac , melanin or neste - coffee led to obtaining 15 - 24 % malformed pupae and 13.0 - 25 % malformed adults. ( Table , 23 ) . These percentages decreased when the spray solutions were left in the field exposed to the natural environmental conditions for 1, 2 and 3 days before offering the treated cotton leaves for larval feeding ( Tables , 24 , 25 & 26 ) . It could be generally deduced from these tables that all the tested materials caused decreased percentages of malformed pupae and adults due to exposure to field conditions ( 2.94 and 3.28 % from exposure of Dipel 2X alone for 3 days; Table, 27 to 10 and 13.64 % from the mixture of the bacterial product flour + yeast + shellac exposed for 24 hours; Table, 25 among pupae and adults, respectively) opposed to 15-24 and 13-25%, respectively % from treatments at zero time; Table, 23. These data indicated that *B. t. kurstaki* showed decreased efficacy in producing malformed individuals by exposure to field conditions and the bad effect of these conditions increased by lengthening the exposure period, on one hand, and that the efficacy of this pathogenic bacteria could be maintained high even after exposure to field conditions to great extent, by adding any of the three used photoprotectants to the mixture of the bioinsecticide + flour + yeast, on the other hand. In all cases, the percentages of malformations were higher adding a photoprotectant than using the bioinsecticide alone or mixed with flour and yeast. But, generally, it could be observed that shellac gave the best results and proved as the best photoprotectant, followed by melanin and neste—coffee, respectively (Table; 24, 25 and 26).

## 3.2.d. Effect of field treatment with bioinsecticide on S. littoralis fecundity and adults' longevity:

The resultant moths from different treatments were paired and allowed to lay their eggs on oleander leaves in glass – chimney cages . The oviposition period , eggs productivity /  $\mathcal{P}$  and males and females' longevity were estimated .

## 3.2.e. Effect on the oviposition period :

Data in Table (27) confirmed what previously deduced in the first part of this study concerning the effect of **B. t. kurstaki** on the oviposition period of **S. littoralis** moth females when the larvae were fed in their first instar on treated food. Treatments of feeding the larvae on treated cotton leaves collected from the field at zero time (

Table (27): Effect of B. thuringiensis kurstaki alone and mixed with photoprotectants on longevity and fecundity of S. littoralis treated at zero time . (Data at 27 C° and 72 % R. H.) .

Treatment	Oviposition	Av. n	Av. no. of	% hatching	Adult's longevity(in days)	evity(in
	(in days)	eggs / female	hatched larvae	0	males	femals
	5.8 ± 0.2 b	248.7 ± 9.0 b	183.3 ± 3.8 b	73 70	6.0 ± 0.3 b	$6.0 \pm 0.3^{\text{ b}}$
B. t. alone	(5.0-6.0)	(233.0-280.0)	(170.0–190.0)	/3./0	(5.0-7.0)	(5.0-7.0)
	5.7 ± 0.2 b	248.3 ± 8.5 b	$180.0 \pm 5.8^{\ b}$	72 40	5.7 ± 0.2 b	$6.0 \pm 0.3^{\ b}$
Flour + yeast + B. L.	(5.0-6.0)	(205.0-290.0)	(170.0-200.0)	12.43	(5.06.0)	(5.0-7.0)
	$5.0 \pm 0.3^{\ b}$	$220.0 \pm 5.8^{\ b}$	$169.0 \pm 13.6^{\ b}$	76 97	$5.3 \pm 0.5^{-6}$	$5.3 \pm 0.5^{\text{ b}}$
Flour + yeast + B. $L + 1\%$ snellac	(4.0-6.0)	(200.0-230.0)	(122.0-195.0)	70.02	(4.0-7.0)	(4.0-7.0)
	5.7 ± 0.2 b	246.7 ± 7.5 b	172.7 ± 7.5 b	70 00	5.7 ± 0.5 b	$6.0 \pm 0.2^{-6}$
Flour + yeast + B. $L + 1\%$ melanin	(5.0-6.0)	(224.0-269.0)	(150.0–195.0)	70.00	(4.0-7.0)	(5.0-7.0)
D 1 1 0/	5.7 ± 0.5 b	$246.7 \pm 13.7^{\text{ b}}$	176.7 ± 6.9 b	71 63	$5.3 \pm 0.2^{-6}$	$6.3 \pm 0.2^{-6}$
Flour + yeast + B. L+ 1 % neste- collec	(4.0-7.0)	(203.0-285.0)	(160.0-200.0)	71.02	(5.0-6.0)	(6.0-7.0)
	10.0 ± 0.3 a	$555.0 \pm 1.7$ a	538.7 ± 5.39 a	07.06	$9.3 \pm 0.2^{-8}$	$10.7 \pm 0.5$ a
Control	(9.0 - 11.0)	(550.0-560.0)	(470.0–598.0)	27.00	(9.0 – 10.0)	(9.0 - 12.0)
F. <sub>(0.05)</sub>	17.75 *	57.23 *	121.95 *		8.08 *	7.25 *
L. S. D. (0.05)	0.99	38.34	26.14		1.43	1.65
					The second secon	THE RESERVE THE PERSON NAMED IN COLUMN TWO IS NOT THE PERSON NAMED IN COLUMN TWO IS NAMED I

just after spraying) caused, significant, decreases in the oviposition period of resultant females ( $5\pm0.3$  days due to feeding on Dipel 2X + flour + yeast + 1 % shellac to  $5.8\pm0.2$  days by feeding on the bioinsecticide alone , opposed to  $10\pm0.3$  days for the untreated females ; Table , 27 ). The differences in oviposition periods between treatments were insignificant . Also , after 24 hours exposure of the assayed materials to field conditions , the resultant female moths from all treatments manifisted , significantly , shorter oviposition period ( $5.3\pm0.5$  to  $6.7\pm0.5$  days ) than the  $10.3\pm0.4$  days recorded from the control females (Table , 28) .

When the sprayed suspension of Dipel 2X alone and its 4 mixtures were left exposed to field conditions after spraying for 48 hours, then offered as contaminated cotton leaves to the first instar S. littoralis larvae, the subsequent adult females showed some prolongation in their oviposition period, indicating decreased efficacy of B. t. kurstaki, although all the recorded periods were also, significantly, shorter (  $6 \pm 0.2 - 8.7 \pm 0.2$  days ) than that of the control (  $10.3 \pm 0.4$ days ) (Table, 29). It could be also noticed from the same table that the oviposition period from using Dipel 2X alone ( $8.7 \pm 0.2$  days) was, significantly, longer than those recorded from the remaining treatments (  $6 \pm 0.2 - 6.7 \pm 0.4$  days; Table, 29). While, by prolongation of the exposure period to 3 days, Dipel 2X alone lost its efficacy on the oviposition period of resultants moths as it lasted 10  $\pm$ 0.3 days being insignificantly different from the  $10.3 \pm 0.4$  days recorded from the control females (Table, 30). As for the remaining treatments those led to longer oviposition periods than in case of 48 hours exposure, although the recorded periods (6.3 - 8.7 days) were sinificantly, shorter than that recorded from the control females.

Table (28): Effect of spraying Dipel 2X alone and mixed with photoprotectants on longevity and fecundity of S. littoralis moths resulting from larvae fed on treated cotton leaves after 24 hours of application .

Treatment	Oviposition	Av. n	Av. no. of	% hatching	Adult's longevity(in days)	evity(in
	(in days)	eggs / female	hatched larvae		males	females
	6.7 ± 0.5 b	340.7 ± 17.5 °	274.7± 16.7°	67.03	$6.7 \pm 0.4^{\text{ b}}$	7.3± 0.5 b
B. t alone	(5.0 - 8.0)	(303.0–358.0) (173.0–273.0)	(173.0-273.0)	80.03	(5.0 - 8.0)	(7.0-9.0)
	6.3 ± 0.2 b	291.7 ± 4.8 b	$210.3 \pm 10.2^{\ b}$	72 00	6.0 ± 0.6 b	$6.7 \pm 0.2^{b}$
Flour + yeast + $\beta$ . $\iota$ .	(6.0 - 7.0)	(275.0-300.0)	(160.0-220.0)	12.07	(4.0-7.0)	(6.0-7.0)
	5.3± 0.5 b	253.3 ± 8.4 b	$170.0 \pm 14.5^{\ b}$	67 11	5.7± 0.4 b	$5.7 \pm 0.2^{b}$
Flour + yeast + $\beta . L + 1\%$ shellac	(4.0-7.0)	(230.0–280.0)	(120.0-200.0)	07.11	(4.0-7.0)	(5.0-6.0)
F1 10/	$6.0 \pm 0.3^{-6}$	$260.3 \pm 11.9^{\text{ b}}$	$180.0 \pm 6.0^{\ b}$	60 15	5.7± 0.8 b	$6.3 \pm 0.2^{-6}$
Flour + yeast + $\beta . L + 1\%$ metanin	(5.0-7.0)	(235.0-301.0)	(165.0-200.0)	07.15	(3.0-7.0)	(6.0-7.0)
T	$6.0 \pm 0.3$ b	268.7 ± 18.3 b	$190.0 \pm 14.2^{\ b}$	70 71	$6.0 \pm 0.3^{-6}$	$6.7 \pm 0.2^{-6}$
Flour + yeast + $B.L+1$ % neste- conee	(5.0-7.0)	(211.0-320.0)	(145.0-230.0)	/0./1	(5.0-7.0)	(6.0-7.0)
	$10.3 \pm 0.4$ a	563.3 ± 1.9 a	549.3 ± 0.2 ª	07 51	$10.0 \pm 0.3$ a	11.0± 0.6 a
Control	(9.0 - 11.0)	(560.0-570.0)	(449.0-500.0)	37.31	(9.0-11.0)	(9.0-12.0)
F. (0.05)	6.19 *	38.90 *	39.19*		7.48 *	8.38 *
L. S. D. (0.05)	1.65	43.16	45.88		2.30	1.40
	SOCIAL PROPERTY OF THE PERSON NAMED AND POST					

Comparing between these 4 treatments , the mixture of flour and yeast to the bioinsecticide gave very limited photoprotection effect than control as the female's average oviposition period lasted  $8.7\pm0.4$  days found , significantly , longer than those recorded from mixtures containing shellac , melanin or neste – coffee ( Table , 30 ) . On the contrary , adding of shellac to the mixture of Dipel 2X + flour and yeast led to highest photoprotection effect as it led after 3 days exposure to the , significantly , shorter oviposition period (  $6.3\pm0.2$  ; 6-7 days ) than all the other treatments ( Table , 30 ) .

## 3.2.f.Effect on eggs' productivity and percentage hatching:

Data in Table (27) show that S. littoralis females that resulted after feeding the first instar larvae on cotton leaves at zero time; i. e. just after field spraying with Dipel 2X or its 4 mixtures suffered significant reductions in the average total number of deposited eggs / female (  $220 \pm 5.8$  eggs from shellac treatment to  $248.7 \pm 9$  eggs from Dipel 2X alone treatment ) than that recorded from the control females (  $555 \pm 1.7 \text{ eggs} / \text{ } \text{?}$  ) . Subsequent reduction occurred in the hatchability percentages among eggs from treatments (70 - 76.82 % ) than control ( 97.1 %). When the spray suspension was left, after application, exposed to environmental conditions for 24 hours before offering the cotton leaves to 1st instar S. littoralis larvae, the resultant female moths deposited also, significantly, lower total number of eggs ( 253.3 - 340.7 eggs / female ) than those deposited by a single female (563.3 eggs; Table, 28). But, it could be noticed control from the mentioned table that those deposited / female from Dipel 2X alone treatment (  $340.7 \pm 17.5 \ \text{eggs}$  ) were , significantly , higher than those recorded from the remaining 4 mixtures 253.3  $\pm$  8.4 – 291.7  $\pm$ 

Table (29): Effect of spraying B. t. kurstaki alone and mixed with photoprotectants on longevity and fecundity of S. littoralis adults resulting from larvae fed on treated cotton leaves after 48 hours of treatment.

Tuotmont	Oviposition	Av. no. of	o. of	% hatching	Adult's longevity(in days)	evity(in
Heatment	(in days)	eggs / female	hatched larvae	0	males	females
	8.7±0.2 b	430.3± 2.5 b	303.7± 1.8 b		$8.0 \pm 0.6^{\ b}$	$10.0 \pm 0.3$ a
B. t. alone	(70 00)	(396.0-410.0)	(301 0-310 0)	82.19	(7.0 - 10.0)	(9.0-11.0)
	(1.0-9.0)	(0.014-0.0/5)	(201.0 210.0)			20.076
	6.7 ± 0.4 °	373.3 ± 10.8 °	$290.0 \pm 3.3$ °	77 68	7.7 ± 0.5 °	8.0 ± 0.7 °
Flour + yeast + $B$ . $t$ .	(6.0 - 8.0)	(290.0-350.0)	(280.0-350.0)	77.00	(6.0 - 9.0)	(6.0-10.0)
	$6.0 \pm 0.2^{\circ}$	282.0 ± <b>4.4</b> °	$171.0 \pm 7.2^{d}$	60 64	5.7± 0.4 °	6.7 ± 0.2 b
Flour + yeast + $B.t.$ + 1% shellac	(5.0-7.0)	(270.0-296.0)	(148.0–191.0)	00.01	(5.0-7.0)	(6.0-7.0)
	6.0 ± 0.2 °	283.3 ± 1.9 °	$203.3 \pm 1.9$ °	71 76	6.0± 0.3°	7.0 ± 0.3 b
Flour + yeast + $B.t.$ + $1\%$ melanin	(6.0-7.0)	(280.0-290.0)	(200.0–210.0)	/1./0	(5.0-7.0)	(6.0-8.0)
	6.7 ± 0.4 °	$312.0 \pm 9.9^{d}$	$228.7 \pm 9.4$ f	73 30	7.3± 0.5 b	7.3 ± 0.5 b
Flour + yeast + $B.t$ + 1 % neste- coffee	(6.0 - 8.0)	(290.0-346.0)	(197.0-238.0)	73.30	(6.0-9.0)	(6.0-9.0)
	10.3 ± 0.4 <sup>a</sup>	558.3 ± 2.5 °	530.7 ± 1.3 <sup>a</sup>	05.00	9.7 ± 0.2 a	$10.7 \pm 0.5^{a}$
Control	(9.0 - 11.0)	(550.0-565.0)	(528.0–535.0)	93.00	(9.0-10.0)	(9.0-12.0)
F. (0.05)	9.3 *	90.09 *	192.9*		4.97 *	8.0 *
L. S. D. (0.05)	1.23	25.42	19.41		1.49	1.30
				And the second s		

4.8 eggs ) which showed , statistically , insignificant differences between them . This increase in eggs' production from Dipel 2X alone treatment indicated the  $\it B.t. kurstaki$  was liable to detrimental effect due to exposure to field conditions , mainly U. V. rays emmited by sunshine , and that the added materials (shellac , melanin and neste – coffee , and even the flour and yeast ) protected , to variable extent , this entomopathogenic bacteria from this effect . This appeared also in the percentages hatching from the obtained eggs which were higher in case of Dipel 2X alone treatment ( 80.6 %) than the remaining 4 treatments ( 67.1 – 72.1 %) , although all of these percentages were lower than that of the control ( 97.5 %, Table , 28 ) .

As the exposure period to field conditions was prolonged to 48 hours, more detrimental effect on B. t. kurstaki occurred, especially in treatment of spraying by Dipel 2X alone as the resultant females laid more eggs , significantly , than the remaining treatments (  $430.3 \pm$ 2.5 eggs / female), although this rate of eggs' production remained, significantly, lower than that produced a single control female (  $558.3 \pm 2.5$  eggs; Table, 29). The percentage hatching was also highest among eggs from females of Dipel 2X alone treatment (82.2) %), but this percentage was also lower than control (95.1 %). Among the remaining treatments, the presence of shellac, melanin or neste - coffee gave protection for the activity of B. t. kurstaki against the environmental conditions, and subsequently the obtained females produced much fewer numbers of eggs (  $282 \pm 4.4$  ,  $283.3 \pm$ 1.9 and 312  $\pm$  9.9 eggs /  $\bigcirc$  , respectively ; Table , 29 ) , although the eggs reproductivity from neste - coffee treatment was significantly, higher than those from the two other treatments. Also, the hatchability percentages from these eggs ( 60.6, 71.8 and 73.3 %, respectively) were much lower than those from treatment of Dipel 2X

Table (30): Effect of spraying Dipel 2 X alone and mixed with photoprotectants on longevity and fecundity of S. littoralis adults resulting from larvae fed on treated cotton leaves after 72 h. of treatment.

Treatment	Oviposition Period	Av. n	Av. no. of	% hatching	Adult's longevity(in days)	evity(in
	(in days)	eggs / female	hatched larvae		males	females
	10.0 ± 0.3 a	515.0± 10.4 a	447.3± 5.4 b	07 05	9.7 ± 0.5 a	$10.3 \pm 0.4^{8}$
B. t. alone	(9.0 - 11.0)	(490.0-550.0)	(430.0-462.0)	86.83	(8.0-11.0)	(9.0-11.0)
	8.7 ± 0.4 b	366.7 ± 7.23 b	320.7 ± 5.3 °	27.70	$9.0\pm0.4$ ad	9.3 ± 0.2 °
Flour + yeast + B. L	(8.0 - 10.0)	(300.0-430.0)	(250.0-380.0)	0/.40	(8.0-10.0)	(9.0-10.0)
	$6.3 \pm 0.2^{d}$	301.0 ± 9.4 b	231.3 ± 10.7 °	70.86	$6.0 \pm 0.3^{-6}$	$7.0 \pm 0.3^{b}$
Flour + yeast + $B.t.$ + 1% shellac	(6.0-7.0)	(280.0-333.0)	(190.0-250.0)	70.00	(5.0-7.0)	(6.0-8.0)
	$7.3 \pm 0.4$ °	$320.0 \pm 6.0^{\ b}$	$240.0 \pm 5.8$ <sup>cd</sup>	75 00	7.0± 0.3 bc	$7.7 \pm 0.5^{-6}$
Flour + yeast + $B.L$ + 1% metanin	(6.0 - 8.0)	(270.0-410.0)	(230.0-260.0)	70:00	(6.0 - 8.0)	(6.0-9.0)
	7.7 ± 0.2 °	$331.0 \pm 11.0^{\ b}$	$259.3 \pm 5.9^{\text{ d}}$	78 34	8.0± 0.3 <sup>cd</sup>	$8.3 \pm 0.4$ b
Flour + yeast + $B.t$ + 1 % neste- coffee	(7.0 - 8.0)	(298.0-364.0)	(250.0-280.0)	70.54	(8.0-10.0)	(7.0-9.0)
	10.3 ± 0.4 a	556.7 ± 1.9 a	530.3± 0.2 <sup>a</sup>	95 06	9.7 ± 0.2 a	$10.7 \pm 0.5$ a
Control	(9.0-11.0)	(550.0-560.0)	(520.0-531.0)	95.00	(9.0-10.0)	(9.0-12.0)
F. (0.05)	17.95 *	13.79 *	41.53*		6.05 *	4.64 *
L. S. D. (0.05)	0.85	66.63	41.28		1.39	1.54
	the state of the last of the l	The same of the sa	-			

alone (82.2%) and that mixed with flour and yeast (77.7%). On the other hand , the average total no of eggs produced by Dipel 2X + flour + yeast treatment (373.3  $\pm$  10.8 eggs /  $\+$ ; Table , 29) was , significantly , higher , than those from the treatments in which either of the three photoprotectants was added to the bioinsecticide .

The detrimental effect of field conditions on the viability of B. t. kurstaki became acute after 3 days exposure in case of spraying by Dipel 2X alone, as, nearly, no effect occurred on the eggs' reproductivity by females that emerged from treated S. littoralis 1st instar larvae. These females produced 515  $\pm$  10.4 eggs /  $\bigcirc$ , being insignificantly lower than the  $556.7 \pm 1.9$  eggs produced by a control female (Table, 30). While on the contrary, addition of, shellac, melanin or neste - coffee kept, to great extent, the activity of the pathogen, and that was clear as a reduction in the average total eggs' productivity / female (  $301 \pm 9.4$  ,  $320 \pm 6$  and  $331 \pm 11$  eggs /  $\mathcal{Q}$  , respectively with insignificant differences between the three treatments; Table, 30). Also, addition of flour and yeast to the bioinsecticide caused some degree of photoprotection as the female deposited a total average of  $366.7 \pm 7.2$  eggs, being insignificantly. higher than the three latter values and, significantly lower than that recorded from Dipel 2X alone treatment. As for the hatchability percentages, those appeared as not greatly affected, although those were higher in treatments of Dipel 2X alone and that mixed with flour and yeast (86.9 and 87.5 %, respectively) and lower in cases of using the mentioned three photoprotectants (70.9, 75 and 78.3%, respectively; Table, 30).

#### 3.2.g. Effect on adults' longevity:

data demonstrated in Table (27), it is clear that the pathogenic effect of Dipel 2X, sprayed on cotton plants for larval treatment, extended in the field up to the adults stage which showed, significantly, shorter life - span of both sexes than control. S. littoralis adults resulted from 1st instar larval feeding on cotton leaves treated in the field, just after spraying of Dipel 2X alone or its mixtures manifested, significantly, shorter longevities (5.3 - 6 days in case of males and 5.3 - 6.3 days in case of females ), being shorter than the 9.3 and 10.7 days, respectively recorded from the control male and female moths ( Table , 27 ). The same significant reduction in moth male and female longevities was also observed after 24 hours exposure of the applied materials to field conditions ( 5.7 - 6.7 and 5.7 - 7.3 days for males and females from treatments, opposed to 10 and 11 days for the control adults, respectively; Table 28). It is clear that the longevities from Dipel 2X alone treatment were, insignificantly, longer (6.7 & 7.3 days ) than those recorded from the remaining 4 treatments (5.7-6 for males and 5.7 - 6.7 days for females; Table, 28) indicating some detrimental effect of the environmental conditions on B. t. kurstaki which manifested some reduction in its efficacy.

More detrimental effect on *B. t. kurstaki* due to exposure to U. V. rays emmited from the sun shine in the field was detected after 48 hours from field application of Dipel 2X alone and that mixed with flour and yeast where lower efficacy was clear on the recorded longevities which lasted 8 & 7.7 days in case of males and 10 & 8 days for females from the two treatments, respectively; Table 29). While, addition of either of the three photoprotectants led to shorter life – spans of emerged moths, being 5.7, 6 & 7.3 days for males and 6.7, 7 & 7.3 days for

females resulted after treatment by shellac, melanin and neste – coffee, respectively ( Table , 29 ) .

The presented data, concerning field application of Dipel 2X, confirm that the main criticism to the field application of **B**. thuringiensis bioinsecticides is the susceptibility of spores of this beneficial group of bacterially to the field conditions, mainly the direct detrimental effect of sunlight UV, causing decreased efficacy even after 24 hours of spraying . This detrimental effect increased, according to the obtained results, by prolongation of the exposure period. In contrast to these results, Abou - Bakr (1997) carried out field applications by spraying Bactospeine EC 13000 IU / ml on cotton plants and he reported that the product showed excellent persistance under field conditions caused 60% mortality among L<sub>1</sub> at 7 days post – treatment. While, in agreement with the present results, Ephraim Cohen et al (1991) reported a rapid loss of B. t. kurstaki toxicity to larvae of Heliothis armigera due to irradiation at 300 - 500 nm for up to 12 hours a photochemical reactor. The same authors indicated that Acriflavin gave the best photoprotection action.

The photoprotection action of shellac , melanin and neste – coffee extended also for 3 days after field spraying of Dipel 2X at the recommended dose on cotton plants , while the bioinsecticide when applied alone or mixed with flour and yeast lost , nearly , all its activity on the longevities of moths reared from treated *S. littoralis* 1<sup>st</sup> instar larvae ( Table , 30 ) . As recorded in the mentioned , table , the males lived for averages of 9.7  $\pm$  0.5 and 9  $\pm$  0.4 days and females lived for 10.3  $\pm$  0.4 and 9.3  $\pm$  0.2 days in cases of application of Dipel 2X or Dipel 2X + flour + yeast , respectively , being insignificantly different

from the 9.7  $\pm$  0.2 and 10.7  $\pm$  0.5 days recorded from the control males and females , respectively . While , on the contrary , moths resulted from larval treatment by Dipel 2X + flour + yeast + either of the three photoprotectants under study lived , significantly , shorter periods than control (  $6\pm0.3$  ,  $7\pm0.3$  days and  $8\pm0.3$  for males and  $7\pm0.3$  ,  $7.7\pm0.5$  and  $8.3\pm0.4$  days for females from the three treatments , respectively ; Table , 30 ) . Thus confirming the excellent photoprotective effect of shellac , melanin and neste – coffee . This effect which kept most of the potentiality of **B. t. kurstaki** even when the spray suspension remained exposed to the action of sun U. V. and other environmental conditions for 3 days .

## 4. MVP $\Pi$ , a genetic engineer product of $\delta$ endotoxin :

## 4.1. Efficacy on mortalities among *S. littoralis* 1<sup>st</sup> and 3<sup>rd</sup> larval instars :

From data in Table (31), it is clear that, 5 days after treatment of 1st and 3rd larval instar of S. littoralis by MVPΠ, the mortality percentages among the treated larvae were a concentration dependent ; i. e., the mortality percentage increased due to increase of the applied concentration. That was clearly evident among the 1st instar larval treatments as the corrected mortality percentages after MVPII treatment ranged from a minimum of 13.3 % by applying the lowest concentration (0.009 ml/100ml water) to a maximum of 96.7 % by using the highest concentration (0.3 ml). The same trend of efficacy was also observed by using the same concentrations on the freshly molted 3<sup>rd</sup> instar larvae (Table, 32) as in the 1<sup>st</sup> instar. After 5 days of starting 3<sup>rd</sup> instar larval feeding on MVPII treated castor - bean leaves, the mortality percentages among the treated larvae ranged from 11 % by using the lowest concentration (0.025 ml) to 93.3 % when the highest concentration (0.8 ml/100 ml. Water) was used ( Table , 32 ). The obtained percentages of mortality, with all concentrations, are lower in case of 3rd instar than those of 1st instar larval treatment ( Tables , 31 & 32 ) indicating a decrease in efficacy as the treated larvae grew older.

Data presented in Table (33) and those illustrated in Fig. (9) indicate that, on the 1<sup>st</sup> instar larvae, the LC<sub>50</sub> after 5 days of MVP $\Pi$  treatment was 0.0388 ml ( with the confidence limites 0.0284 and 0.0518 ml at 95 % confidence limits ). While, by treatment of the

Table (31): Corrected mortality percentages among S. littoralis  $1^{st}$  instar larvae fed on castor – bean leaves treated with MVP  $\Pi$  (values after 5 days of treatment).

Concentration (ml / 100ml water)	Means of corrected mortality %
0.009	13.3
0.018	33.3
0.037	46.7
0.075	66.7
0.15	80.0
0.3	96.7

Table (32): Corrected mortality percentages among  $3^{rd}$  larval instar of **S.** littoralis fed on leaves treated with MVP  $\Pi$  (values after 5 days of treatment).

Concentration (ml / 100ml water)	Means of corrected mortality %
0.025	11.0
0.05	30.0
0.1	40.0
0.2	53.3
0.4	73.3
0.8	93.3

Table (33): LC<sub>50</sub> and LC<sub>90</sub> values after 5 days of S. littoralis 1<sup>st</sup> and 3<sup>rd</sup> instars larval treatment with MVPII.

Treated	Slope	$\mathrm{LC}_{50}$	95 % confidence limits	dence limits	$\mathrm{LC}_{90}$	95 % confidence limits	lence limits
indtar	,		lower	npper		lower	upper
1 <sup>st</sup>	1.6822	0.0388	0.0284	0.0518	0.2243	0.1467	0.4389
3 <sup>rd</sup>	1.6009	0.1405	0.1033	0.1910	0.8880	0.5509	1.9346

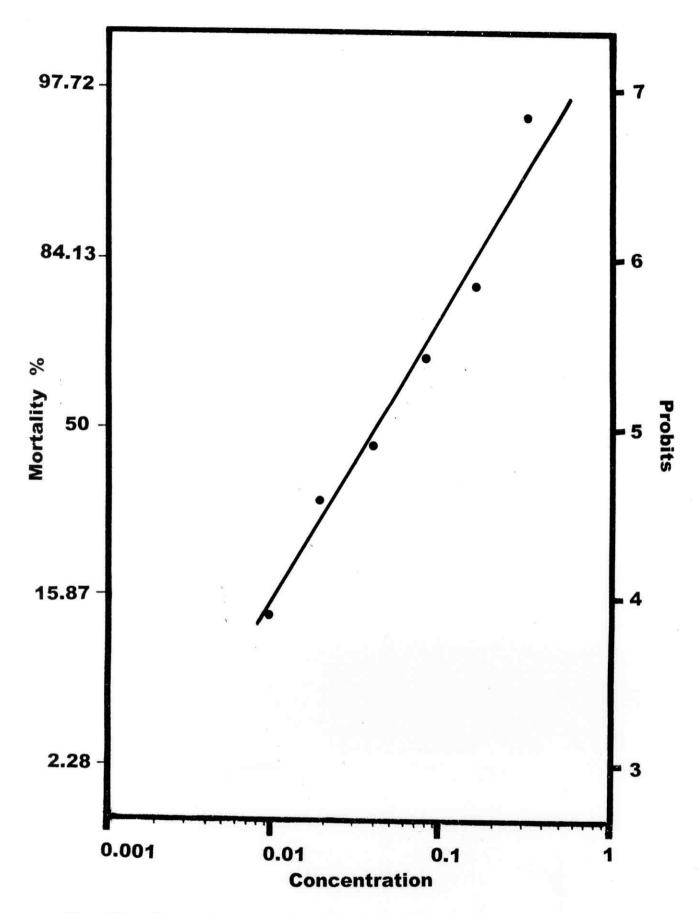


Fig. (9): Regression mortality lines after of *S. littoralis* larvae fed in their  $1^{st}$  larval instar for 5 days on castor – bean leaves treated with MVP $\Pi$ .

third instar larvae , the LC  $_{50}$  after 5 days of MVP $\Pi$  treatment was 0.1405 ml (0.1033 – 0.1910 ml) (Table , 33 and Fig. 10). These data proved a positive correlation between the age of larvae at the time of treatment and the needed concentration of the biocide in order to reach the LC  $_{50}$  level .

## 4.2. Effect of S. littoralis $1^{st}$ and $3^{rd}$ instar larval treatments by the LC $_{50}$ of MVP $\Pi$ on the survivors :

The 1<sup>st</sup> & 3<sup>rd</sup> instar larvae were fed for 2 days on treated castor – bean leaf discs by the LC<sub>50</sub> of MVP $\Pi$  (0.0388 ml), then the survivors were allowed to feed on clean untreated castor – bean leaves until pupation and subsequently adults' emergence. The experiment was carried out under constant conditions of 27  $\pm$  1 C°& 60 – 65 % R. H.

### Effect on larval and pupal durations :

As shown in Tables ( 34 & 35 ),  $1^{st}$  and  $3^{rd}$  instar larval treatments by the LC  $_{50}$  of MVPII ( 0.0388 and 0.1405 ml ) caused significant elongations in the larval and pupal periods . These periods lasted  $17.8 \pm 0.4$  and  $11.4 \pm 0.2$  days , among treated larvae and pupal , respectively , opposed to  $11.6 \pm 0.2$  and  $8.2 \pm 0.3$  days , respectively among the control individuals when treatment occurred on the  $1^{st}$  instar larvae ( Table , 34 ) . While , by treatment on the  $3^{rd}$  instar , these durations lased  $15.4 \pm 0.4$  and  $10.8 \pm 0.3$  , respectively for treatment opposed to  $9.8 \pm 0.3$  and  $8.6 \pm 0.3$  days for control ( Table , 35 ) .

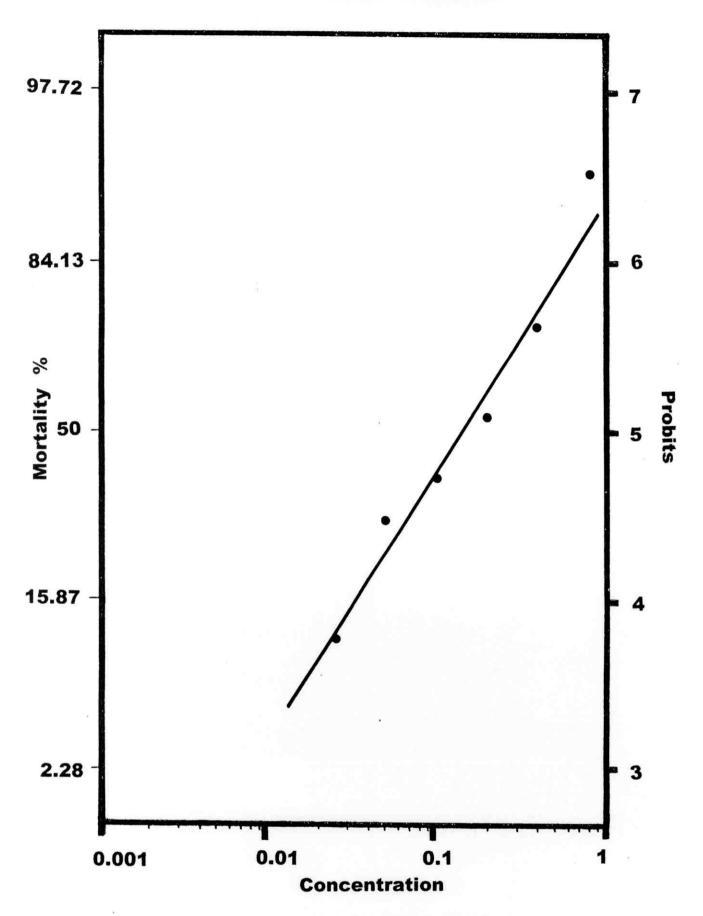


Fig. (10): Regression lines showing mortality percentages of S. *littoralis* among the larvae fed in their  $3^{rd}$  treated with MVP $\Pi$ .

Table (34): Treatment of 1st instar S. littoralis larvae by LC50 of MVP II in relation to larval and pupal periods and larvae at the beginning of experiment). mortality percentages amongst larvae, prepupae and pupae (data from 100 freshly emerged 1st instar

		4.85*	7.04*					T. (9.85)
0.00	0.00	± 0.2 8.2 ± 0.3 13.0) (7.0 - 10.0)	$11.6 \pm 0.2$ $(11.0 - 13.0)$	2.08	2.04	2.00	0.00	Control
18.75	7.25	$17.8 \pm 0.4 \qquad 11.4 \pm 0.2$ $(16.0 - 20.0) \qquad (10.0 - 12.0)$	$17.8 \pm 0.4$ $(16.0 - 20.0)$	17.65	20.00	52.50	0.0383	ПАЛМ
adults	pupae	pupae	larvae	pupae	prepupae	larvae	(ml/100ml water)	Пеасшен
rmations	% malformations	Durations ( in days )	Durations	ong	% mortality among	% п	Concentration	Treatment

Table (35): Treatment of 3rd instar S. littoralis larvae by LC50 of MVP II in relation to larval and pupal periods and mortality percentages amongst larvae, prepupae and pupae (data from 100 freshly emerged 3rd instar larvae at the beginning of experiment).

Treatment	Concentration	% п	% mortality among	ong	Durations	Durations ( in days )	% malformations	mations
	(mb/100ml water)	larvae	prepupae	pupae	larvae	pupae	pupae	adults
MVPII	0.1405	56.00	10.71	16.00	$15.4 \pm 0.4$ $10.8 \pm 0.3$ $(13.0 - 18.0)$ $(9.0 - 12.0)$	10.8 ± 0.3 18.0) (9.0 - 12.0)	9.52	15.79
Control	0.00	2.00	0.00	2.04	$9.8 \pm 0.3$ $8.6 \pm 0.3$ $(9.0 - 12.0)$ $(7.0 - 10.0)$	$8.6 \pm 0.3$ $(7.0 - 10.0)$	0.00	0.00
T. (0.05)					4.44*	2.65		

#### Larval, prepupal and pupal mortalities:

When *S. littoralis*  $1^{st}$  and  $3^{rd}$  instar larvae were fed for 48 hours on castor – bean leaf discs treated with the LC  $_{50}$  of MVP $\Pi$ , followed by feeding on untreated castor – bean leaves, the mortality percentages among the treated larvae reached 52.5 and 56 % for the two instars , respectively, opposed to 2 % among the control larvae (Tables , 34 & 35 ). As for the subsequent prepupae , mortalities were also detected and recorded . Those were estimated being 20 and 10.7 % for those developed from treated  $1^{st}$  and  $3^{rd}$  instar , respectively , opposed to 2 and 0.0 % among the prepupae that developed from untreated larvae . The pupae that developed from the surviving prepupae showed also mortalities . Those comprised 17.65 % for those developed from  $1^{st}$  instar larval treatment ( Table , 34 ) and 16 % among those developed from  $3^{rd}$  instar larval treatment ( Table , 35 ) , opposed to 2.08 and 2.04 % among pupae that developed from the control larvae .

### • Pupae and adults' malformation:

No malformations could be detected among the pupae and adults developed from the control treatments. While, among those developed from MVPII treated larvae their were malformations which could be detected by eye inspection (Fig. 11). The deformations of pupae and moths were, nearly, similar to those previously reported in case of Dipel 2X treatments (s.p. 60). The malformation percentages were 7.25 and 18.75% among pupae and moths resulted after 1st instar larval treatment (Table, 34), and 9.52 and 15.79%, respectively among

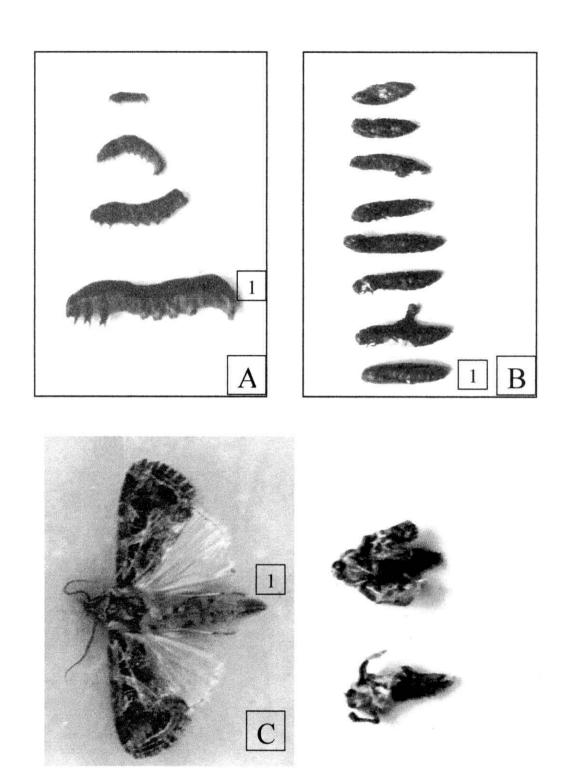


Fig. (11): Malformed S. littoralis stages after larval treatment with  $MVP\Pi$ .

- A. Normal (1) and stunted larvae .
- B. Normal (1) and malformed pupae. C. Normal (1) and malformed adults.

pupae and adults , respectively , developed after  $3^{\text{rd}}$  instar larval treatment ( Table , 35 ) .

#### ◆ Effect of MVP ☐ larval treatment on moths fecundity:

Resultant moths after S. *littoralis* 1<sup>st</sup> and 3<sup>rd</sup> instars larval treatment, by MVP $\Pi$  at the LC<sub>50</sub> concentration, were allowed to deposit their eggs on oleander leaves placed in glass chimney cages. The averages of oviposition period, total no of eggs / female, % hatching among the deposited eggs and the male & female longevity were recorded.

From data presented in Tables ( 36 & 37 ), feeding of the  $1^{\rm st}$  or  $3^{\rm rd}$  instar larvae of  $\bf S$ . *littoralis* on castor – bean leaves treated by MVPII caused significant reductions in the oviposition period and total productivity of eggs deposited by a subsequent female than control . The oviposition period lasted  $6.7 \pm 0.3$  ( 6.0 - 8.0 ) days for females resulted from the first instar larval treatment ( Table , 36 ) and  $7.3 \pm 0.2$  ( 7.0 - 8.0)days for those emerged after  $3^{\rm rd}$  instar larval treatment ( Table , 37 ), opposed to  $10.3 \pm 0.2$  ( 10.0 - 11.0 ) days in case of the control female . Throughout the oviposition period the total number of deposited eggs averaged  $318.3 \pm 35.2$  ( 300.0 - 340.0 ) and  $338.0 \pm 18.4$  ( 330.0 - 344.0 ) eggs / female resulted from treatment of the  $1^{\rm st}$  and  $3^{\rm rd}$  larval instars , respectively . While , a control female of the two experiments deposited  $595 \pm 6$  and  $594.3 \pm 6.0$  eggs, respectively (Tables , 36 & 37) .

The percentages of hatching of S. *littoralis* eggs were also found to be reduced due to larval feeding on castor – bean leaves treated by the endotoxin , being 81.68 % from eggs deposited by females resulted after 1<sup>st</sup> instar larval treatment ( Table , 36 ) and 85.89 % from eggs deposited after rearing the larvae treated in their 3<sup>rd</sup> instar by MVPII ( Table , 37 )

Table (36): Effect of S. littoralis 1st instar larval treatment by MVP  $\Pi$  on fecundity and longevity of the resultat adults.

Treatment	Concentration	Oviposition	Av. I	Av. no of	% hatching	Adult's longevity ( in days )	ity ( in days )
Licaemone	(mV100 ml water)	Period (in days)	eggs / 💠	hatched larvae	g	99	<del>\$</del> \$
МVРП	0.0388	$6.7 \pm 0.3$ $(6.0 - 8.0)$	$318.3 \pm 35.2$ $260.0 \pm 37.0$ $(300.0 - 340.0)$ $(253.0 - 313.0)$	$260.0 \pm 37.0$ (253.0 -313.0)	81.68	$7.3 \pm 0.6$ $(6.0 - 9.0)$	$7.5 \pm 0.5$ $(6.0 - 9.0)$
Control	0.00	$10.3 \pm 0.2$ $(10.0 - 11.0)$	$595.0 \pm 6.0 \qquad 587.3 \pm 5.6$ $(580.0 - 615.0) \qquad (573.0 - 606.0)$	$587.3 \pm 5.6$ (573.0 - 606.0)	98.70	$9.9 \pm 0.2$ $(9.0 - 10.0)$	$10.9 \pm 0.2$ (10.0 - 11.0)
T. <sub>(0.05)</sub>		5.00*	17.98*	14.71*		5.90*	5.15*

. These percentages were 98.7 and 98.2 % from eggs deposited by the control female from the two experiments , respectively ( Tables ,36 & 37 ) .

#### ◆ Effect of MVP∏ larval treatment on adults' longevity :

Male and female moths developed after feeding the  $1^{st}$  or  $3^{rd}$  instar larvae on castor – bean leaves , treated by the LC  $_{50}$  of MVP\Pi , lived for significantly , shorter periods than those recorded for moths developed from rearing of the untreated larvae . That is clear from Tables ( 36~&~37 ) which show that male and female moths lived for  $7.3\pm0.6$ and  $7.5\pm0.5$  days , respectively when the preceding  $1^{st}$  instar larvae were treated by the LC50 of MVPH , while treatment of the  $3^{rd}$  instar larvae by the endotoxin led to male and female moths lived for  $7.1\pm0.1$  and  $8.1\pm0.3$  days , respectively , opposed to the averages of 9.9 and 10.9 days as life – spans for the males and females developed from the control larvae .

From the aforementioned results concerning the efficacy of MVPII, it could be concluded that treatment of 1<sup>st</sup> or 3<sup>rd</sup> instars of *S. littoralis* by the endotoxin caused considerable mortality levels, which were a concentration dependent, among the treated larvae and subsequent prepupae and pupae. Among the survivors, malformed pupae and adults were detected, while the resultant adults manifested, shorter oviposition period through which females deposited fewer numbers of eggs from which lower hatchability percentages were recorded, and also the means of male and female longevities were shorter than those recorded from the control moths.

Table (37): Effect of S. littoralis  $3^{rd}$  instar larval treatment by MVP  $\Pi$  on fecundity and longevity of the resultat adults.

	Concentration	Oviposition	Av. no of	10 of	% hatching	Adult's longevity ( in days )	ity ( in days )
Treatment	(ml/100 ml water)	Period (in days)	eggs / 💠	hatched larvae		0,	+0 +0
MVP II	0.1405	$7.3 \pm 0.2$ $(7.0 - 8.0)$	$338.0 \pm 18.4$ $290.0 \pm 18.7$ $(330.0 - 344.0)$ $(283.0 - 324.0)$	$290.0 \pm 18.7$ (283.0 -324.0)	85.89	$7.1 \pm 0.1$ (7.0 - 8.0)	$8.1 \pm 0.3$ $(8.0 - 10.0)$
Control	0.00	$10.3 \pm 0.2$ $(10.0 - 11.0)$	$594.3 \pm 6.0$ $583.7 \pm 5.1$ $(588.0 - 615.0)$ $(570.0 - 600.0)$	583.7 ± 5.1 (570.0 - 600.0)	98.22	$9.9 \pm 0.3$ $(8.0 - 10.0)$	$10.9 \pm 0.1$ $(10.0 - 11.0)$
T. (0.05)		6.50*	29.36*	24.32*		8.96*	5.70*

### 4.3. Effect of MVP $\Pi$ exposure to UV:

#### 4.3.a. Laboratory experiment:

Data presented in Table (38) present the mortality percentages due to *S. littoralis* 1<sup>st</sup> instar larval feeding on castor – bean leaves treated with MVPII at concentration of 0.3 ml in 100 ml. water for 2 days, then the larvae were fed on untreated leaves for another 3 days. The endotoxin solution was used without any radiation treatment, and after exposure to U V radiation for 16, 36, 44 and 58 hours. It is clear from the obtained results that the unexposed material caused the highest mortality percentage among the treated larvae (96.7%). The efficacy of MVPII decreased due to exposure to U V light, and the mortality percentage, among treated larvae, decreased as the exposure period to U V became longer. The lowest mortality percentage (46.7%) occurred due to exposure of the endotoxin to U V for the longest period (58 hours). This percentage increased to 70,76.7 and 93.3% as the exposure period was shortened to 44,36 and 16 hours, respectively.

### 4.3.b. Field experiment:

This experiment was carried out by spraying both sides of the leaves of three castor – bean plants by the MVP $\Pi$  solution at 0.3 ml / 100 ml water. Castor – bean leaves were collected from the treated trees and offered , in the laboratory , to **S. littoralis** 1<sup>st</sup> instar larvae for a couple of days after which the larvae were fed on fresh untreated leaves . Collection of treated leaves from the field took place at zero time (just after spraying), then after 2, 4, 9, 11 and 14 days.

Table (38): Means of larval mortality % among S. littoralis fed on castor - bean leaves treated with UV irradiated MVPΠ after 5 days of treatment.

Exposure period (hours)	Larval mortality %
0.0	96.7
16	93.3
36	76.7
44	70.0
58	46.7

Table (39): Means of larval mortality % among S. littoralis in their  $1^{\text{st}}$  instar larvae fed on castor – bean leaves treated with UV irradiated MVP  $\Pi$  after 5 days of treatment.

oosure Mean of corrected		Means	
	C°	R. H.	
93.3	18	50	
86.7	20	47	
80.0	21	49	
60.0	20	50	
50.0	20	51	
36.7	21	50	
	86.7 80.0 60.0 50.0	mortality % after 5 days       C°         93.3       18         86.7       20         80.0       21         60.0       20         50.0       20	

As shown in Table ( 3.9 ), the highest mortality percentage ( 93.3% ) occurred when the offered treated castor – bean leaves were collected just after treatment ( before being exposed to sunlight or any other environmental conditions ). This percentage decreased to 86.7% when the sprayed solution of MVPII was left exposed to natural environmental conditions ( especially sunlight U V ) for two days . On the other hand , the lowest mortality percentage among treated larvae reached 36.7% when the exposure period in the field was lengthened to the maximum period of 14 days . By using the treated leaves after 4 ,9 and 11 days exposure , to natural environmental conditions , before being offered to the  $1^{\rm st}$  instar of *S. littoralis* larvae , the mortality percentages among the treated larvae after 5 days of treatment reached 80 , 60 and 50%, respectively .

Both the laboratory and field data obtained after exposure to U V radiation ( in the laboratory ) and all the environmental conditions including sunlight U V ( in the field ) confirm that MVPΠ (*B.t.* δ endotoxin ) was resistant to the direct effect of U V irradiation even when exposed continuously to U V for 36 hours in the laboratory , as the endotoxin caused 76.7 % mortality of the treated larvae . Also , exposure of the bioinsecticide to field conditions continuously for 4 days led to 80 % mortality among the treated larvae , the percentage which could be considered as high because it occurred after 4 days exposure . These results indicate that MVPΠ which is a product of the δ endotoxin *B. t. kurstaki* bioencapsulated in killed *Pseudomonas fluorescens* is more tolerant to the detrimental effect of the UV irradiation that the common commercial bioinsecticides which contain the *B. thuringiensis* spores , as in the latter case it was found the Dipel 2X (*B. t. kurstaki* spores) lost about 45 % of its activity after 24 hours exposure to field conditions

(mortality of *S. littoralis* larvae decreased from 65 % when the  $1^{\rm st}$  instar larvae were fed on treated cotton leaves to 36 % when treatment took place after 24 hours of spraying, Table, 23 & 24).

## 5. Haemolymph studies:

## 5.1. Determination of total haemocyte counts (THCs )in

## S. littoralis larvae treated with Dipel 2X:

Haemolymph of *S. littoralis* 2<sup>nd</sup> and 4<sup>th</sup> instar larvae was drawn up to 0.5 mark in a thoma of white blood cell dilution pipette and diluted with turek's solution for blood counting.

As shown in Table (40) and Fig. (12), the total number of haemocytes decreased gradually by increasing the concentrations of Dipel 2X as compared with control (  $30583 \pm 83.43$  cells ); being  $21566 \pm 136.58$  and  $15316 \pm 585.45$  cells when larvae were treated in their  $2^{nd}$  instar and  $25733 \pm 259.04$  and  $18550 \pm 293.32$  haemocyte cells by treatment of the 4th instar with LC20 and LC50 of Dipel 2X, respectively. The obtained results showed also that more effect of the bioinsecticide in reducing the total haemocytes count occurred by treatment of larvae in their earlier instar (2nd instar) than treatment of the older instar ( 4th instar ). In the former instar, the reduction percentages due to treatment on the LC 20 and LC 50 of Dipel 2X were 29.48 and 49.92 %, respectively. While, by treatment of the 4th instar larvae, these percentages of reduction were 15.86 and 39.34 %, respectively than control (Table, 40). These rates of reduction in the total haemocyte counts were, statistically, significant than control in all treatment. It is clear from the mentioned table, that the severest effect of Dipel 2X on the total haemocyte count occurred when the 2<sup>nd</sup> instar S. littoralis larvae were fed on leaves treated by the LC50 of the bioinsecticide (  $15316 \pm 585$  ; 14500 - 16450 cells ).

The present results concerning the effect of feeding **S.** *littoralis* larvae on treated leaves with **B.** *thuringiensis* in reducing the THCs in

Table(40): Effect of Dipel 2X treatment on the total haemocyte counts per mm<sup>3</sup> blood of S. littoralis 2 nd and 4th instar larvae (counts after 120 h. treatment).

Concentration (I. U./mg)	Larval instar treated	Mean ± S.E and range	Reduction %
Control		(a) $30583 \pm 83.43$ (30500 - 30750)	-
LC <sub>20</sub> 0.3300 X 10 <sup>3</sup>	2 <sup>nd</sup>	(b) $21566 \pm 136.58$ $(21300 - 21750)$	- 29.48
0.4900 X 10 <sup>3</sup>	4 <sup>th</sup>	(c) $25733 \pm 259.04$ $(25500 - 26250)$	- 15.86
LC <sub>50</sub> 1.0592 X 10 <sup>3</sup> 1.8432 X 10 <sup>3</sup>	2 <sup>nd</sup>	(d) $15316 \pm 585.45$ (14500 - 16450) (e) $18550 \pm 293.32$ (18000 - 19000)	- 49.92 -39.34
L.S.D. (0.05)		850.85	

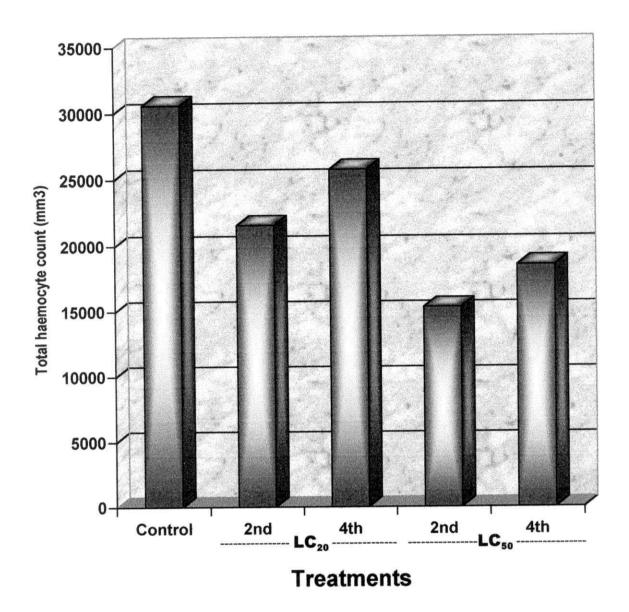


Fig. (12): Total haemocyte counts (THCs) of the 2<sup>nd</sup> and 4<sup>th</sup> instar of S. littoralis larvae treated with LC<sub>20</sub> and LC<sub>50</sub> of Dipel 2X.

the larval haemolymph are in harmony with those previously recorded by *Rosemberger* and *Jones (1960)*, *Vankova & Leskova (1972)*, *Gagen* and *Ratcliffe (1976)* and *El – Mandrawy (1992)*, who indicated that the THCs of insects vary greatly with technique, species, stages of development, various physiological conditions within each stage and concentration of the applied *B. thuringiensis*.

### 5.2. Types of haemocytes:

On basis of light microscopy inspections, the eight types of haemocytes on the haemolymph of *S. littoralis* larvae (Fig. 13 to 20) were nearly the same eight types previously described by *Abd El-Rahman* (1994) and Kares (1994).

# 5.3. Determination of the differential haemocyte counts ( DHCs ) :

## 5.3.1. Qualitative analysis of haemocytes of *S. littoralis* larvae :

#### Prohaemocytes:

Table (41) shows , the averages of nucleus dimensions of the prohaemocyte cells for the untreated second instar of *S. littoralis* larvae were  $5.7\pm0.33$ ,  $5.7\pm0.33$  &  $6.0\pm0.33$   $\mu$  in length and  $5.5\pm0.33$  5.6  $\pm$  0.01 &  $5.2\pm$  0.17  $\mu$  in width while the averages of cell dimensions were  $6.7\pm0.33$  ,  $6.8\pm0.17$  &  $6.7\pm0.33$   $\mu$  in length and  $6.7\pm0.33$  ,  $6.8\pm0.17$  &  $6.7\pm0.33$   $\mu$  in width after 72 , 96 and 120 hours ; respectively .

Table (41): Effect of Dipel 2X treatment on the biometric measurements of Prohaemocytes in treated 2<sup>nd</sup> and 4<sup>th</sup> instar larvae of S. littoralis

				Length (µ	( <del>प</del>				
3	Cell	Cell (h. after treatment	ent)	Nucleu	Nucleus ( h. after treatment	ment)	Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment)	er treatment)
1 reatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>									
Control	$6.7 \pm 0.33$	$6.8 \pm 0.17$	$6.7 \pm 0.33$	$5.7 \pm 0.33$	$5.7 \pm 0.33$	$6.0 \pm 0.33$	85.07	83.82	89.55
$LC_{20}$	$6.7 \pm 0.44$	$6.3 \pm 0.03$	$6.0\pm0.01$	4. 7± 0.33	$4.0\pm0.03$	$3.8 \pm 0.17$	70.15	63.49	54.29
$LC_{50}$	$7.0 \pm 0.01$	$6.2 \pm 0.29$	$5.3 \pm 0.07$	$4.0 \pm 0.00$	$3.0 \pm 0.00$	$2.7 \pm 0.03$	57.14	48.38	50.94
4 <sup>th</sup>									
Control	$7.7 \pm 0.33$	$7.3 \pm 0.29$	$7.7 \pm 0.03$	$6.4 \pm 0.00$	$6.3 \pm 0.30$	$6.7 \pm 0.33$	83.12	86.30	87.01
$LC_{20}$	$7.3 \pm 0.33$	$7.0 \pm 0.01$	$7.0 \pm 0.03$	$5.7 \pm 0.33$	$4.7 \pm 0.33$	$4.7 \pm 0.37$	78.08	67.14	67.14
$LC_{50}$	$7.0 \pm 0.44$	$7.0 \pm 0.03$	$6.3 \pm 0.03$	$4.3 \pm 0.33$	$3.7 \pm 0.33$	$3.3 \pm 0.17$	58.90	52.86	52.38
L.S.D. between con.	$2^{nd} = 0.8042$		$4^{th} = 0.7857$	$2^{nd} = 0.7474$		$4^{th} = 0.9234$			
between time	$2^{nd} = N. S.$		$4^{th} = N. S.$	$2^{nd} = N. S.$		$4^{th} = N. S.$			
Interaction	$2^{nd} = N. S.$		4th = N.S.	$2^{nd} = N.S.$		4th = N.S.			
				Width (µ)	(μ)				
2 <sup>nd</sup>									
Control	$6.7 \pm 0.33$	$6.8 \pm 0.33$	$6.7 \pm 0.33$	$5.5 \pm 0.33$	$5.6 \pm 0.01$	$5.2 \pm 0.17$	84.62	82.35	83.58
$LC_{20}$	$6.0\pm0.01$	$6.2 \pm 0.29$	$6.0 \pm 0.33$	$3.7 \pm 066$	$3.5 \pm 0.29$	$3.0 \pm 0.01$	60.77	53.85	47.62
$LC_{50}$	$5.3 \pm 0.03$	$5.0 \pm 0.03$	$5.0 \pm 0.00$	$3.3 \pm 0.21$	$3.0 \pm 0.01$	$2.7 \pm 0.33$	49.25	50.00	50.94
4 <sup>th</sup>				, a					
Control	$7.0 \pm 0.00$	$7.0 \pm 0.33$	$7.0 \pm 0.01$	$6.0 \pm 0.00$	$6.0 \pm 0.01$	$6.0 \pm 0.01$	85.71	88.19	85.19
$LC_{20}$	$6.3 \pm 0.33$	$6.3 \pm 0.58$	$6.3 \pm 0.33$	$5.0 \pm 0.00$	$4.0 \pm 0.00$	$4.7 \pm 0.03$	79.36	66.67	74.60
$LC_{50}$	$5.7 \pm 0.03$	$5.3 \pm 0.33$	$5.0 \pm 0.00$	$4.0 \pm 0.00$	$3.7 \pm 0.33$	$3.3 \pm 0.44$	70.17	69.81	66.00
L.S.D. between con.	$2^{nd} = 0.7067$		$4^{th} = 0.9234$	$2^{nd} = 0.9849$		$4^{th} = 0.5687$			
between time	$2^{nd} = N. S.$		4th = N.S.	$2^{nd} = N.S.$		4th = N.S.			
Interaction	$2^{nd} = N. S.$		$4^{th} = N.S.$	$2^{nd} = N. S.$		4 <sup>th</sup> = N.S.			

**Treatment** Control LC 20 LC<sub>50</sub> 72 h. 2<sup>nd</sup> larval instar after 96 h. 120 h. 0.03 mm72 h. 4th larval instar after 96 h. 120 h.

Fig. (13): Prohaemocytes cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

On 4<sup>th</sup> instar larvae , the averages of nucleus dimensions were 6.4 , 6.3  $\pm$  0.30 & 6.7  $\pm$  0.33  $\,\mu$  in length and 6.0 , 6.0  $\pm$  0.01 & 6.0  $\,\mu$  in width while the averages of cell dimensions were 7.7  $\pm$  0.33 , 7.3  $\pm$  0.29 & 7.7  $\pm$  0.03  $\,\mu$  in length and 7.0 , 7.3  $\pm$  0.33 & 7.0  $\pm$  0.01  $\,\mu$  in width after 72 , 96 and 120 hours , respectively .

Also , the nucleus cell ratios were 85.07, 83.82 & 89.55 % for length measurements and 84.62, 82.35 & 83.58 % for width measurements after 72, 96 and 120 hours , respectively .

When *S. littoralis*  $2^{nd}$  and  $4^{th}$  instar larvae were fed on castor – bean leaves contaminated with  $LC_{20}$  and  $LC_{50}$  of Dipel 2X , these treatments led to decreases in prohaemocyte cell and nucleus dimensions , and also in the nucleus cell ratios ( Table , 41) . These decreases were more pronounced by using the higher concentration ( $LC_{50}$ ), on one hand , and also by lengthening the period after treatment . Also , the prohaemocyte cells and nuclei took irrigular shapes , compared to those of control (Fig. 13).

### Plasmatocytes :

The dimensions of the plasmatocyte cell of the healthy larvae averaged 14.7  $\pm$  0.70 , 13.7  $\pm$ 1.20 & 14.7  $\pm$  0.67  $\mu$  in length and 13.3  $\pm$  0.67 , 13.0  $\pm$  1.00 & 13.7  $\pm$  0.67  $\mu$  in width, and 15.7  $\pm$  0.66 , 15.7  $\pm$  0.88 & 14.3  $\pm$  1.2  $\mu$  long and 14  $\pm$  1.16 , 14.3  $\pm$  0.88 & 12.7  $\pm$  0.88  $\mu$  wide for the second instar after 72 , 96 and 120 hours , respectively. While those of the nucleus were 7.3  $\pm$  0.30 , 6.7  $\pm$  0.33 & 6.7  $\pm$  0.60  $\mu$  in length and 6.7  $\pm$  1.03 , 6.0 & 6.3  $\pm$  0.33  $\mu$  in width for the second instar and 8  $\pm$  0.5 , 8.8  $\pm$  0.53 & 8  $\pm$  0.58  $\mu$  long and 7.7  $\pm$  0.7 , 8  $\pm$  0.03 & 6.7  $\pm$  0.52  $\mu$  wide for the 4th instar after 72 , 96 and 120 hours ,

Table (42): Effect of Dipel 2X treatment to S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae on the biometric measurements of Plasmatocytes.

				Length (μ)	(μ)				
	Cell	Cell (h. after treatment	ent)	Nucleu	Nucleus (h. after treatment )	ment)	Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment	er treatment)
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>									
Control	$14.7 \pm 0.70$	13.7± 1.20	$14.7 \pm 0.67$	$7.3 \pm 0.30$	$6.7 \pm 0.33$	$6.7 \pm 0.60$	49.66	48.90	45.58
LC <sub>20</sub>	$13.3 \pm 0.33$	$12.7 \pm 033$	$11.7 \pm 0.33$	$5.0 \pm 0.76$	$5.3 \pm 0.33$	$5.3 \pm 0.33$	37.59	41.73	45.29
LC <sub>50</sub>	$12.7 \pm 0.33$	$11.0 \pm 0.00$	$10.7 \pm 0.33$	$4.7 \pm 0.30$	$4.3 \pm 0.17$	$4.0 \pm 0.58$	37.00	39.09	37.38
4 <sup>th</sup>									
Control	15.7 ± 0.66	$15.7 \pm 0.88$	$14.3 \pm 1.20$	$8.0 \pm 0.50$	$8.8 \pm 0.53$	$8.0 \pm 0.58$	50.95	56.05	55.94
LC <sub>20</sub>	$14.7 \pm 0.33$	$13.3 \pm 0.33$	$13.0 \pm 0.58$	$6.2 \pm 0.44$	$6.3 \pm 0.30$	$6.3 \pm 0.17$	42.18	47.37	48.46
LC <sub>50</sub>	$13.7 \pm 0.33$	$10.7 \pm 0.70$	$10.3 \pm 0.65$	$5.0 \pm 0.01$	$4.0 \pm 0.00$	$3.8 \pm 0.20$	36.49	37.38	36.89
I S D between con	$2^{nd} = 1.6887$		$4^{th} = 2.0360$	$2^{nd} = 1.3717$		4th = N.S.			54).
between time	11		$4^{th} = 2.0360$	$2^{nd} = 1.3717$		11			
Interaction	$2^{nd} = N.S.$		$4^{th} = N. S.$	$2^{nd} = N. S.$		4th = N.S.			
				Width (μ)	(H)				
2 <sup>nd</sup>									
Control	$13.3 \pm 0.67$	$13.0 \pm 1.00$	$13.7 \pm 0.67$	$6.7 \pm 1.03$	$6.0 \pm 0.00$	$6.3 \pm 0.33$	50.37	46.15	45.98
$LC_{20}$	$12.0 \pm 0.00$	$11.0 \pm 0.58$	$10.7 \pm 0.33$	$5.5 \pm 0.29$	$4.9 \pm 0.17$	$5.3 \pm 0.33$	45.33	44.54	49.53
LC <sub>50</sub>	$11.3 \pm 0.33$	$10.0 \pm 0.33$	$9.7 \pm 0.00$	$4.0 \pm 0.67$	$3.5 \pm 0.29$	3.3± 0.17	35.39	35.00	34.02
4 <sup>th</sup>									
Control	$14.0 \pm 1.16$	$14.3 \pm 0.88$	$12.7 \pm 0.88$	$7.7 \pm 0.70$	$8.0 \pm 0.03$	$6.7 \pm 0.52$	55.00	55.94	54.33
$LC_{20}$	$13.0 \pm 0.67$	$12.0 \pm 0.58$	$11.0\pm0.00$	$5.8 \pm 0.38$	$5.3 \pm 0.17$	$5.0 \pm 0.58$	44.61	44.17	45.45
LC <sub>50</sub>	$11.7 \pm 0.33$	$10.0 \pm 0.58$	$9.9 \pm 0.90$	$3.3 \pm 0.33$	$3.0 \pm 0.58$	+	28.20	30.00	30.30
L.S.D. between con.	$2^{nd} = 1.5808$		$4^{th} = 6.7090$	$2^{nd} = 1.38824$		11			
between time	$2^{nd} = N.S.$		$4^{th} = 6.7090$	$2^{nd} = N. S.$		4th = N.S.			
Interaction	$2^{nd} = N. S.$		$4^{th} = N. S.$	$2^{nd} = N. S.$		4 <sup>th</sup> = N.S.			
STREET, STREET	STREET, STREET	STREET, STREET		THE RESIDENCE AND ADDRESS OF THE PERSON NAMED IN COLUMN TWO IS NOT THE OWNER.		The second secon	The second secon		

Control Treatment 72 h. larval instar after 96 h. 120 h. 0.03 mm72 h. larval instar after 96 h. 120 h.

Fig. (14): Plasmatocytes cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

respectively . The nucleus cell ratios are 49.66, 48.40 and 45.58 in the second instar and 50.37, 46.15 and 45.98 in the fourth instar after 72, 96 and 120 hours, respectively (Table, 42).

By Dipel 2X treatment to  $2^{nd}$  and  $4^{th}$  instar larvae, the plasmatocyte cell and nucleus dimensions diminished, and also the nucleus cell ratio decreased ( Table , 42 ). Some morphological changes could be also detected. Those included the nucleus which lost its central position and moved towards the cell wall. As occurred with the plasmatocytes, the cells took irregular shape, compared to those from the haemolymph of control larvae ( Fig. 15 ).

#### Granular cells :

The average granular cell dimensions of healthy  $2^{nd}$  and  $4^{th}$  instar larvae were  $10.7\pm0.33$ ,  $10.3\pm0.33$  &  $11.3\pm0.33$   $\mu$  in length and 10.0, 10.0 and  $10.3\pm0.33$  in width for the  $2^{nd}$  instar  $11.7\pm$ ,  $11.7\pm0.33$   $\mu$  long and  $11.7\pm0.33$  & 11  $\mu$  wide for the  $4^{th}$  instar after 72, 96 and 120 hours , respectively .While , those of the nuclei measured  $9.5\pm0.43$  ,  $9.7\pm0.24$  &  $9.9\pm0.33$   $\mu$  in length and  $9.4\pm0.51$  , 9.6 and  $9.3\pm0.24$   $\mu$  in width for the  $2^{nd}$  instar , and 10  $\mu$  long and 10 ,  $9.9\pm0.03$  &  $9.7\pm0.33$   $\mu$  wide for the fourth one after 72 , 96 and 120 hours , respectively (Table , 43).The nuclear cell ratios in the  $2^{nd}$  instar were 88.78 , 94.17 and 87.61 % for length and 94.00, 96.80 and 90.29% for width while those in the  $4^{th}$  instar were 85.47 , 85.47 & 85.87% for length and 85.47 , 84.61 & 88.18% for width after 72 96 and 120 hours , respectively . Concerning the effect of larval feeding on Dipel 2X treated food on the granular cells , Fig . (15) shows that cells changed to be of smaller size and irregular

Table (43): Effect of Dipel 2X treatment on the biometric measurements of Granular cells in S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

				Length (u	(m)				
	Call	Cell (h after treatment	nt)	Nucleu	Nucleus ( h. after treatment	ment)	Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment)	er treatment)
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>									
Control	$10.7 \pm 0.33$	10.3 ±0.33	$11.3 \pm 0.33$	$9.5 \pm 0.43$	$9.7 \pm 0.24$	$9.9 \pm 0.33$	88.78	94.17	87.61
IC.	$10.0 \pm 0.00$	$9.7 \pm 0.03$	$9.3 \pm 0.33$	$8.3 \pm 0.17$	$5.3 \pm 0.65$	$5.0 \pm 0.29$	83.00	54.64	33.63
IC.	9.3 ± 0.01	$9.0 \pm 0.00$		$6.3 \pm 0.30$	$4.7 \pm 0.33$	$4.0 \pm 0.33$	67.74	52.22	45.98
4th	- 1								
Control	$11.7 \pm 0.33$	$11.7 \pm 0.33$	$11.7 \pm 1.33$	$10.0 \pm 0.00$	$10.0 \pm 0.00$	$10.0 \pm 0.00$	85.47	85.4/	85.87
IC	107 + 033	$10.3 \pm 0.88$	$10.0 \pm 0.03$	$7.3 \pm 0.33$	$7.0 \pm 0.29$	$6.5 \pm 0.67$	68.22	67.96	65.00
10.9	97 ±0.35	9.7 ± 0.29	$9.0 \pm 0.03$	$7.3 \pm 0.70$	$6.3 \pm 0.30$	$4.7 \pm 0.70$	75.26	64.95	52.22
	$2^{nd} = 0.8043$		$4^{th} = 1.5479$	11		$4^{th} = 1.2946$			
between time	ıı		4th = N.S.	$2^{nd} = N. S.$		$4^{th} = 1.2946$			
Interaction	н		$4^{th} = N.S.$	$2^{nd} = N.S.$		4 <sup>th</sup> = N.S.			
				Width (μ)	(μ)				
2 <sup>nd</sup>								200	20.20
Control	$10.0 \pm 0.00$	$10.0 \pm 0.00$	$10.3 \pm 0.33$	$9.4 \pm 0.51$	$9.6 \pm 0.00$	$9.3 \pm 0.24$	94.00	96.80	90.29
I.C.	$9.7 \pm 0.33$	8.3 ± 0.33	$8.7 \pm 0.33$	$7.3 \pm 0.35$	$5.0 \pm 0.29$	$5.0 \pm 0.29$	75.26	60.24	60.24
LC <sub>50</sub>	- 1	- 1	$7.7 \pm 0.00$	$6.0 \pm 0.29$	$4.7 \pm 0.33$	$3.9 \pm 0.33$	72.29	58.75	50.65
4 <sup>th</sup>									00 10
Control	$11.7 \pm 0.33$	$11.7 \pm 0.33$	$11.0 \pm 0.00$	$10.0 \pm 0.00$	$9.9 \pm 0.03$	$9.7 \pm 0.33$	85.47	84.61	88.18
LCm	$10.3 \pm 0.33$	$9.7 \pm 0.58$	$9.7 \pm 1.00$	$7.3 \pm 0.37$	$6.0 \pm 0.29$	$5.5 \pm 0.58$	70.87	61.85	36.70
LCs	$8.7 \pm 0.33$	8.3 ± 0.29	$8.0 \pm 0.66$	$6.0 \pm 0.27$	$5.0 \pm 0.29$	5 ±	68.96	60.24	56.25
1 6 D between 200	11		$4^{th} = 0.8743$	$2^{nd} = 0.9699$		11.			
between time	n		н	$2^{nd} = 0.9699$		$4^{th} = 1.2002$			
	11		$4^{th} = N.S.$	$2^{nd} = N. S.$		4 <sup>th</sup> = N.S.			

LC<sub>50</sub> **Treatment** Control 72 h. 2<sup>nd</sup> larval instar after 96 h. 120 h. 0.03 mm 72 h. larval instar after 96 h. 120 h.

Fig. (15): Granular cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

wall due to treatment, while the nuclei the tended to leave their central position and move towards the cell margins.

### · Spindle cells:

The averages of dimensions of spindle cells from healthy 2<sup>nd</sup> and 4<sup>th</sup> instar larvae of **S.** littoralis measured  $15.7 \pm 1.20$ ,  $15.7 \pm 1.20$ & 15.7  $\pm$  1.76  $\mu$  in length and 11.7  $\pm$  1.67, 12.0  $\pm$  1.53 & 11.7  $\pm$  $1.67\mu$  in width for the former instar , and  $19.7\pm0.88$  ,  $19\pm1.16$  &  $20.3 \pm 0.33 \mu \text{ long}$ , and  $11.6 \pm 0.88$ ,  $10.7 \pm 0.67$  and  $12 \pm 0.58$  wide  $\mu$  in the latter one after 72 , 96 and 120 hours , respectively . While those of nuclei were  $8.7 \pm 0.53$ ,  $9.0 \pm 1.00$  &  $8.9 \pm 0.81$   $\mu$  in length and  $7.3 \pm 0.67$  ,  $6.7 \pm 0.63~\&~6.5 \pm 0.59~\mu$  in width for the  $2^{\text{nd}}$  instar , and 8.3  $\pm~0.67$  , 8.8  $\pm~0.15$  & 9  $\pm~0.29$   $\mu$  long and 7.1  $\pm~0.6$  , 6.7  $\pm$  $0.88~\&~8.3\pm0.33~\mu$  wide for the  $4^{th}$  instar . The nuclear cell ratios in length were estimated by 55.41, 57.32 & 56.69 % in the 2<sup>nd</sup> insar and 42.13, 46.31 & 44.33 in the 4th instar after 72 96 and 120 h., respectively, (Table, 44). Concerning the effect of larval feeding on Dipel 2X treated food on spindle cells, Table (44) shows a decrease in the cell dimensions and more occurred by increase the Dipel 2X concentration and by prolongation of the period after treatment, although the effect of increasing bioinsecticidal concentration was more pronounced. While, Fig. (16) shows clearly the decrease in cell dimensions and the changes in the cell shape due to treatment.

Table (44): Effect of Dipel 2X treatment on the biometric measurements of Spindle cells in S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

				Length (µ	(μ)			. (0/ ) ( 1 .4.	
	Cell	Cell (h. after treatment	ent)	Nucleus	s (h. after treatment )		Nucleus cell Rat	Nucleus cell Ratio (%) ( n. after treatment	er treatment
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>								20,00	09.75
Control	$15.7 \pm 1.20$	$15.7 \pm 1.20$	$15.7 \pm 1.76$	$8.7 \pm 0.53$	$9.0 \pm 1.00$	8.9 ± 0.81	55.41	37.32	30.09
I.C.	$15.7 \pm 0.67$	$14.0 \pm 0.58$	$13.0 \pm 0.58$	$8.7 \pm 0.67$	$8.7 \pm 0.88$	8.0 ± 0.33	55.41	62.14	62.05
I.C.	$13.3 \pm 0.33$	$12.0 \pm 0.58$	$11.7 \pm 1.86$	$7.0 \pm 0.00$	$6.3 \pm 0.33$	$6.3 \pm 0.33$	62.63	72.50	33.83
4 <sup>th</sup>			N.					16.21	11 22
Control	$19.7 \pm 0.88$	$19.0 \pm 1.16$	$20.3 \pm 0.33$	$8.3 \pm 0.67$	$8.8 \pm 0.15$	$9.0 \pm 0.29$	42.13	46.31	51 07
IC.	$17.0 \pm 0.58$	$15.7 \pm 0.33$	$16.0 \pm 0.58$	$8.0 \pm 0.58$	$8.3 \pm 0.33$	8.3 ± 0.67	47.06	52.87	50.00
1020	157+067	$15.3 \pm 0.33$	$14.0 \pm 1.00$	$8.3 \pm 0.33$	$7.7 \pm 0.33$	$7.0 \pm 0.67$	52.87	30.33	00.00
LC 50	$2^{\text{nd}} = 2.6783$		$4^{th} = 2.1139$	$2^{nd} = 4.3003$		11			
L.S.D. between con.	11		II	$2^{nd} = N. S.$		11			
Interaction	11		4th = N.S.	$2^{nd} = N. S.$		4"" = N.S.			
				Width (μ)	(μ)				
2 <sup>nd</sup>							(2.20	20.02	77.75
Control	$11.7 \pm 1.67$	$12.0 \pm 1.53$	$11.7 \pm 1.67$	$7.3 \pm 0.67$	$6.7 \pm 0.63$	6.5 ± 0.59	62.39	33.63	\$8,00
LCm	$11.0 \pm 0.33$	$11.0 \pm 0.33$	$10.0 \pm 0.00$	$7.0 \pm 0.58$	$7.3 \pm 0.33$	5.8 ± 0.90	03.04	72.70	50.00
LC <sub>50</sub>	$7.7 \pm 0.33$	$7.3 \pm 0.33$	$6.7 \pm 0.33$	$5.3 \pm 0.33$	$5.3 \pm 0.33$	4.0 ± 0.00	68.00	72.00	33.70
4 <sup>th</sup>						62-022	61 21	63 63	69 17
Control	$11.6 \pm 0.88$	$10.7 \pm 0.67$	$12.0 \pm 0.58$	1.1 ± 0.00	0./±0.00	0.0 + 0.00	20.00	70 97	70 87
LC	$11.0 \pm 0.58$	$10.3 \pm 0.58$	$10.3 \pm 0.58$	$7.7 \pm 0.67$	7.3 ± 0.70	/.3 ± 0.33	/0.00	70.07	61.20
I.C.	$9.0 \pm 0.33$	$8.7 \pm 0.88$	$8.7 \pm 0.33$	.7 ±	$6.0 \pm 0.58$	$5.7 \pm 0.33$	63.33	08.90	01.29
1 5 7				$2^{\text{nd}} = 1.4749$		n			
between time	II		$4^{th} = N. S.$	$2^{nd} = 1.4749$		11			
Interaction	ij		4th = N.S.	$2^{nd} = N. S.$		4" = N.S.			

Control Treatment LC 20  $LC_{50}$ 72 h. 2<sup>nd</sup> larval instar after 96 h. 120 h. 0.03 mm 72 h. 4<sup>th</sup> larval instar after 96 h. 120 h.

Fig. (16): Spindle cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

### Adipohaemocytes:

The dimensions of adipohaemocytic cells , nuclei and nucleus cell ratios from the haemolymph of healthy and Dipel 2X treated S. *littoralis*  $2^{nd}$  and  $4^{th}$  instar larvae are recorded in Table (45). The presented data show that treatments by the  $LC_{20}$  caused ,in many cases slight decrease in the length and or width of cells of the  $2^{nd}$  instar larvae , while decrease in cell and nucleus dimensions could be observed by increasing the applied dose to the  $LC_{50}$  to both the  $2^{nd}$  and  $4^{th}$  instar , but all the differences due to treatments were , statistically insignificant . Also the statistical values showing the interaction between concentration and time after treatment were insignificant . Drawings of the adipohaemocytes from healthy and treated larvae are given in Fig. (17). This figure may demonstrate the decrease in size due to treatment , but no characteristic morphological changes in the cell walls and the nucleus shape and position could be observed .

### Oenocyte cells :

Feeding of the *S. littoralis*  $2^{nd}$  and  $4^{th}$  instar larvae on castor – bean leaves treated with the  $LC_{20}$  of Dipel 2X caused slight decrease in the oenocyte cell length and width especially after 96 and 120 hours after treatment . More decrease in the cell dimensions occurred by raising the bioinsecticide concentration to the  $LC_{50}$  level , but these decreases were also insignificant , compared to oenocyte cells of the untreated larvae . While , the nuclei of oenocytes from larvae treated with the  $LC_{20}$  of Dipel 2X showed , in some cases , significant decrease in length and width than control , while in most cases the  $LC_{50}$  treatment caused ,

Table (45): Effect of Dipel 2X treatment on the biometric measurements of Adipohaemocytes in S. littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

				Length (μ	(μ)				
	Cell	Cell (h. after treatment	ent)	Nucleus	ıs (h. after treatment	ment)	Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment)	er treatment)
Treatment	72	96	120	72		120	72	96	120
2 <sup>nd</sup>									
Control	$18.7 \pm 2.14$	$19.3 \pm 2.97$	$18.7 \pm 1.45$	$3.3 \pm 0.33$	$3.5 \pm 0.01$	$3.3 \pm 0.33$	17.64	18.13	17.65
$LC_{20}$	$18.0 \pm 2.08$	$19.3 \pm 0.67$	$16.7 \pm 2.02$	$2.7 \pm 0.33$	$3.3 \pm 0.33$	$3.3 \pm 0.33$	15.00	17.09	19.76
LC <sub>50</sub>	$19.7 \pm 3.72$	$19.0 \pm 1.00$	$18.3 \pm 1.67$	$3.0 \pm 0.00$	$3.0 \pm 0.03$	$3.0 \pm 0.00$	15.23	15.79	16.39
4 <sup>th</sup>									
Control	$21.7 \pm 3.33$	$20.0 \pm 2.89$	$22.0 \pm 1.76$	$4.0\pm0.00$	$3.7 \pm 0.33$	$3.3 \pm 0.33$	18.43	18.50	15.00
$LC_{20}$	$13.3 \pm 0.88$	$17.6 \pm 1.86$	$17.3 \pm 1.67$	$3.0 \pm 0.00$	$3.7 \pm 0.67$	$3.0 \pm 0.58$	16.39	21.02	17.34
LC <sub>50</sub>	$18.0 \pm 1.00$	$18.0 \pm 0.58$	$19.0 \pm 2.51$	$3.3 \pm 0.33$	$3.3 \pm 0.33$	$3.0 \pm 0.58$	18.33	18.33	15.79
L.S.D between con	$2^{nd} = N.S.$		$4^{th} = N.S.$	$2^{nd} = N. S.$		$4^{th} = N.S.$			
between time	11		11	$2^{nd} = N. S.$		$4^{th} = N.S.$			
Interaction	$2^{nd} = N. S.$		$4^{th} = N. S.$	$2^{nd} = N.S.$		4th = N.S.			
				Width (μ)	(H)				
2 <sup>nd</sup>									
Control	$15.7 \pm 0.33$	$16.0 \pm 1.16$	$16.3 \pm 1.33$	$2.7 \pm 0.67$	$3.0 \pm 0.00$	$2.7 \pm 0.33$	17.19	18.75	16.56
$LC_{20}$	$16.0 \pm 2.31$	$12.3 \pm 1.86$	$14.0 \pm 1.53$	$2.0 \pm 0.00$	$2.0 \pm 0.01$	$2.0 \pm 0.00$	12.50	16.26	14.28
LC <sub>50</sub>	$10.3 \pm 0.33$	$13.3 \pm 1.67$	$13.3 \pm 1.67$	$2.0 \pm 0.00$	$2.3 \pm 0.03$	$2.3 \pm 0.33$	19.41	17.29	17.29
4 <sup>th</sup>									
Control	$15.0 \pm 0.00$	$18.3 \pm 1.98$	$18.0 \pm 1.16$	$3.3 \pm 0.33$	$2.7 \pm 0.33$	$2.7 \pm 0.33$	22.00	14.75	15.00
$LC_{20}$	$16.7 \pm 0.88$	$16.0 \pm 1.53$	$14.0\pm1.00$	$2.3 \pm 0.33$	$3.3 \pm 0.33$	$3.3 \pm 0.00$	13.77	23.57	14.28
LC <sub>50</sub>	$14.0 \pm 1.00$	$16.7 \pm 0.88$	$12.7 \pm 2.51$	$2.7 \pm 0.33$	$3.0 \pm 0.01$	$3.0 \pm 0.58$	19.28	17.96	23.62
L.S.D. between con	$2^{nd} = N.S.$		$4^{th} = N. S.$	$2^{nd} = N. S.$		4th = N.S.			
between time	н		$4^{th} = N. S.$	$2^{nd} = N. S.$		4th = N.S.			
Interaction	$2^{nd} = N. S.$		4 <sup>th</sup> = N. S.	$2^{nd} = N. S.$		$4^{th} = N. S.$			
					THE REAL PROPERTY AND PERSONS ASSESSED.				

LC 20 Control **Treatment** 72 h. larval instar after 96 h. 0.03 mm 72 h. larval instar after 96 h. 120 h.

Fig. (17): Adipohaemocytes cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

significant, decrease in the dimensions of nuclei (Table, 46). Also, the nucleus cell ratio decreased due to treatments, and the decrease was more pronounced by increasing the concentration, on one hand, and prolongation of the period after treatment, on the other hand (Table, 46).

As shown in Fig. (18), few changes in the morphological shape of oenocyte may by observed due to bioinsecticidal treatment such as losing the normal spherical shape of the cell, being attenuated or rectangular or losing the central position of the nucleus

### · Spherule cells:

The effect of Dipel 2X treatment on spherule cells from the haemolymph of  $2^{nd}$  and  $4^{th}$  *S. littoralis* larval instars was more pronounced than that occurred in cases of adipohaemocytes and oenocytes . As shown in Table ( 47 ) the  $LC_{20}$  treatments caused detectable decrease in the cells and nuclei dimensions even after 72 h. from treatment of both instars , and this decrease became , significantly , different than that recorded from control larvae after 96 hours from treatment . While , using the higher concentration (  $LC_{50}$  ) caused , significant , decrease in cells and nuclei dimensions even after 72 hours from treatment . These results indicate the high sensitivity of the spherule cell to bioinsecticidal treatments . Also , decreased nuclear – cell ratio could be detected , and more decease occurred by raising the used concentration of the bioinsecticide under investigation .

Fig. (19) shows drawings of spherule cells from haemolymph of healthy and Dipel 2X treated larvae. Changes due to treatments included irregularity in the outline of the cell and nucleus walls and, in some cells, the tendency of nuclei to leave its central position and move towards the cell wall.

Table (46): Effect of Dipel 2X treatment on the biometric measurements of Oenocyte cells in S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

				Length (µ	(µ)				
	Call	Call (h after treatment	nt)	Nucleu	Nucleus (h. after treatment	ment)	Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment)	er treatment)
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>									
Control	$10.0 \pm 0.01$	$10.7 \pm 0.58$	$10.0 \pm 0.55$	$5.3 \pm 0.33$	$5.7 \pm 0.33$	$5.4 \pm 0.21$	53.00	53.27	54.00
10.3	$10.0 \pm 0.58$	$9.7 \pm 0.33$	$9.7 \pm 0.33$	$4.7 \pm 0.33$	$4.6 \pm 0.65$	$4.3 \pm 0.35$	47.00	47.42	44.33
I.C.	9.0 ± 0.58	8.3 ± 0.33	$8.0 \pm 0.46$	$4.0 \pm 0.29$	$3.7 \pm 0.17$	$4.3 \pm 0.33$	44.44	51.81	58.75
4th									
Control	$11.3 \pm 0.33$	$11.7 \pm 1.05$	$11.7 \pm 0.33$	$5.3 \pm 0.17$	$6.3 \pm 0.33$	$6.0 \pm 0.29$	46.90	53.85	21.28
1.03	$11.3 \pm 1.20$	$10.0 \pm 0.00$	$10.0 \pm 0.00$	$5.3 \pm 0.65$	$4.7 \pm 0.33$	$4.3 \pm 0.35$	46.90	47.00	43.00
I.C.	$10.3 \pm 0.33$	$9.7 \pm 0.33$	$9.0 \pm 0.00$	$4.5 \pm 0.29$	$4.0 \pm 0.29$	.5 ±	43.69	41.24	38.89
1 6 1 1	$2^{nd} = 1.26$		$4^{th} = 1.6176$	$2^{nd} = 1.0979$		H			
between time	11		$4^{th} = N.S.$	$2^{nd} = N. S.$		4" = N.S.			
Interaction	$2^{nd} = N.S.$		4th = N.S.	$2^{nd} = N.S.$		4" = N.S.			
				Width (μ)	(μ)				
2 <sup>nd</sup>								25.53	
Control	$9.7 \pm 0.33$	$9.3 \pm 0.75$	$9.3 \pm 0.88$	$5.0 \pm 0.00$	$5.0 \pm 0.00$	$5.5 \pm 0.33$	51.55	33.76	39.14
LCm	$9.7 \pm 2.61$	$8.7 \pm 0.58$	$8.7 \pm 1.22$	$4.0 \pm 0.00$	$4.3 \pm 0.06$	$4.0 \pm 0.29$	41.24	49.42	45.98
LC <sub>50</sub>	$8.3 \pm 0.33$	$8.0 \pm 0.00$	$7.7 \pm 0.33$	$4.6 \pm 0.58$	$3.3 \pm 0.17$	$3.0 \pm 0.00$	48.19	41.25	38.96
4 <sup>th</sup>									
Control	$10.3 \pm 1.47$	$10.7 \pm 0.73$	$10.3 \pm 1.20$	$5.3 \pm 0.33$	$5.3 \pm 0.33$	$5.3 \pm 0.33$	51.46	49.53	31.46
10	$10.0 \pm 1.00$	$9.0 \pm 0.00$	$9.0 \pm 0.29$	$5.0 \pm 0.76$	$4.3 \pm 0.33$	$4.0 \pm 0.29$	50.00	47.78	44.44
LCso	9.7 ± 0.33	$9.0 \pm 0.69$	$8.3 \pm 0.33$	$4.3 \pm 0.29$	$3.7 \pm 0.33$	3 ±	44.33	41.11	39.76
1 6 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1		- 4	$4^{th} = N.S.$	$2^{nd} = 0.1212$		$4^{th} = 1.26$			
between time	11		4th = N.S.	$2^{nd} = N. S.$		II			
Interaction	11		4th = N.S.	$2^{nd} = N.S.$		4 <sup>th</sup> = N.S.			

	LC <sub>50</sub>	LC 20	Control	Treatment	
				72 h.	2 <sup>nd</sup> 1
				96 h.	2 <sup>nd</sup> larval instar after
0.03			×	120 h.	•
03 mm				72 h.	4 <sup>th</sup> 1:
				96 h.	4 <sup>th</sup> larval instar after
				120 h.	er

Fig. (18): Oenocyte cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

Table (47): Effect of Dipel 2X treatment on the biometric measurements of Spherule cells in S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

		,		Length (μ)	h after	treatment)	Nucleus cell Rat	Nucleus cell Ratio (% ) (h. after treatment	r treatment)
	Cell (	Cell (h. after treatment	nt)	Nucieus	-n. alter		3	20	130
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>							21 02	51 02	53 00
Control	$11.0 \pm 0.58$	$11.0 \pm 0.58$	$11.3 \pm 1.20$	$5.7 \pm 0.67$	$5.7 \pm 0.67$	6.0 ± 0.58	20.12	20.10	16.22
C*	$9.7 \pm 0.30$	$9.7 \pm 0.33$	$9.3 \pm 1.20$	$4.7 \pm 0.33$	$4.3 \pm 0.33$	$4.3 \pm 0.33$	48.45	44.32	20.75
1020		$9.3 \pm 0.33$	$8.3 \pm 0.60$	$4.3 \pm 0.32$	$4.3 \pm 0.33$	$3.3 \pm 0.33$	44.32	46.24	39.73
4th	- 1	- 1							
	117+033	120+000	$11.7 \pm 0.33$	$6.7 \pm 0.33$	$7.0 \pm 0.00$	$6.7 \pm 0.33$	57.26	38.33	37.20
Control	107 1022	10.0 + 0.00	93 +033	$6.0 \pm 0.57$	$5.3 \pm 0.33$	$5.3 \pm 0.30$	56.07	53.00	56.90
LC20	-	00 + 0.57	- 1	$5.0 \pm 0.00$	$4.7 \pm 0.33$	$4.3 \pm 0.29$	53.76	52.22	51.18
LC50	and - 20647	7.0 - 0.0 .	11 1	$2^{nd} = 1.3609$		$4^{th} = 1.0288$			
L.S.D. between con.	II		11	$2^{nd} = N. S.$		11			
Interaction	11		4th = N.S.	$2^{nd} = N.S.$		4" = N.S.			
				Width (µ)	(μ)				
2 <sup>nd</sup>				50.050	52±033	53+033	50.00	50.00	50.00
Control	$10.0 \pm 0.00$	$10.3 \pm 0.33$	10	3.0 ± 0.30		3 3 + 0 33	44 44	44.44	39.76
$LC_{20}$	$9.0 \pm 0.57$	9.0 ± 0.00	1	4.0 ± 0.00	1.0 + 0.07	20.0 + 0.00	41 11	37.93	35.00
$LC_{50}$	9.0 ± 0.50	8.7 ± 0.00	8.0 ± 0.23	J. / ± 0.00	0.0				
4 <sup>th</sup>		107.000	110+023	62+033	63±0.33	$6.3 \pm 1.73$	60.30	58.88	57.27
Control	8C.0 ± 0.11		07 1022	57+067	50+057	43±0.33	57.00	51.55	49.42
LC <sub>20</sub>	$10.0 \pm 0.58$	1.	8.7 ± 0.55	3.7 + 0.01	10+000	36+033	45.98	48.19	46.75
	$8.7 \pm 0.33$	$8.3 \pm 0.33$	$7.7 \pm 0.00$	± 0.	4.0 ± 0.00	1	10.50		
ED between con	$2^{nd} = 1.2245$		$4^{th} = 1.1374$	$2^{nd} = 1.8861$		11			
between time	11		4th = N.S.	2 <sup>nd</sup> = N.S.		11			
Interaction	II		$4^{th} = N. S.$	$2^{nd} = N. S.$		4" = N.S.			

 $LC_{50}$ LC 20 Control **Treatment** 72 h. 2<sup>nd</sup> larval instar after 96 h. 120 h. 0.03 mm 72 h. larval instar after 96 h. 120 h.

Fig. (19): Spherule cell types recognized in untreated and Dipel 2X treated S. littoralis larvae

### Cystocyte cells:

dimensions of a cystocyte from the averages cell haemolymph untreated  $2^{nd}$  and  $4^{th}$  larval instar were  $10.7 \pm 0.33$ , 11.0and 10.0  $\pm$  0.33  $\mu$  in length and 10.0 , 10.3  $\pm$  0.33 & 10.3  $\pm$  0.33  $\mu$  in width for the former instar and 11.7  $\pm~0.69$  , 12  $\pm~0.58$  & 12.3  $\pm~0.88~\mu$ long and 11.7  $\pm~0.33$  , 10.7  $\pm~0.33$  & 11.3  $\pm~0.88~\mu$  wide for the latter one . Those of a nucleus of the  $2^{nd}$  instar larvae were  $5.8\pm0.55$  ,  $6.3\pm$ 0.40~ and  $~5.0~\pm~0.58~\mu$  in length and  $5.7\pm0.69$  ,  $5.3\pm0.17~$  &  $5.0~\mu~$  in width and in the 4<sup>th</sup> instar were  $6.3 \pm 0.65$ ,  $6.3 \pm 0.35$  &  $6.7 \pm 0.53$   $\mu$ long and 5.3  $\pm~0.33$  , 5.3  $\pm~0.33$  & 5.5  $\pm~0.15~\mu$  wide after 72 , 96 and 120 h., respectively. Accordingly, the nuclear - cell ratios were 54.20 , 57.27 & 46.73 % for length and 57.00 51.46 and 48.54 % for width in the 2<sup>nd</sup> instar and 53.85, 52.5 & 54.47 for length and 49.53, 49.53 & 48.67 for width in the 4th instar after 72, 96 and 120 h., respectively (Table  $\,$ , 48) . When larvae were fed on leaves treated with LC<sub>20</sub> and LC<sub>50</sub> of Dipel 2X, the cell dimensions decreased and the rate of decrease increased by increasing the bioinsecticidal concentration to LC50, on one hand, and prolongation of the period after treatment, on the other hand ( Table , 48 ) . These decreases in cell dimensions could be easily observed in the illustrated drawings (Fig. 20), in addition to the of cells and nuclei, and also the tendency of the nucleus to irrigularity leave its position.

Table (48): Effect of Dipel 2X treatment on the biometric measurements of Cystocyte cells in S.littoralis 2<sup>nd</sup> and 4<sup>th</sup> instar larvae.

				Length (μ	(μ)			. (0/ )/ L .#	
	Cell	Cell (h. after treatment	ent)	Nucleus	s (h. after treatment		Nucleus cell Ra	Nucleus cell Ratio (%) (h. after treatment	er treatment
Treatment	72	96	120	72	96	120	72	96	120
2 <sup>nd</sup>									
Control	$10.7 \pm 0.33$	$11.0 \pm 0.00$	$10.7 \pm 0.33$	$5.8 \pm 0.55$	$6.3 \pm 0.40$	$5.0 \pm 0.58$	54.20	57.27	46./3
LC	$9.7 \pm 0.33$	$9.7 \pm 0.33$	$9.7 \pm 0.33$	$4.7 \pm 0.33$	$4.3 \pm 0.33$	$4.0 \pm 0.00$	47.92	44.33	41.12
LCsa		- 1		$4.3 \pm 0.33$	$4.0 \pm 0.00$	$3.3 \pm 0.33$	46.24	44.00	42.86
4 <sup>th</sup>	- 1								
Control	$11.7 \pm 0.69$	$12.0 \pm 0.58$	$12.3 \pm 0.88$	$6.3 \pm 0.65$	$6.3 \pm 0.35$	$6.7 \pm 0.53$	53.85	52.50	34.47
LC	$10.7 \pm 0.33$	$10.3 \pm 0.33$	$10.3 \pm 0.33$	$5.3 \pm 0.88$	$5.3 \pm 0.33$	$5.0 \pm 0.01$	49.53	51.46	48.53
ICa	$9.7 \pm 0.58$	$9.0 \pm 0.33$	$8.9 \pm 0.06$	$4.3 \pm 0.33$	$4.3 \pm 0.33$	4.0± 0.29	44.33	47.78	44.94
1 6 50	11 [	- 1	$4^{th} = 1.6712$	$2^{nd} = 1.0979$	2	11			
between time	11		н	$2^{nd} = 1.0979$		4 <sup>th</sup> = N.S.			
Interaction	11		4 <sup>th</sup> = N.S.	$2^{nd} = N. S.$		4" = N.S.			
				Width (µ	(μ)				
2 <sup>nd</sup>									1051
Control	$10.0 \pm 0.00$	$10.3 \pm 0.33$	$10.3 \pm 0.33$	$5.7 \pm 0.69$	$5.3 \pm 0.17$	$5.0 \pm 0.00$	57.00	51.46	48.54
LCm	$8.7 \pm 0.33$	$8.7 \pm 0.67$	$8.3 \pm 0.33$	$4.7 \pm 0.33$	$4.3 \pm 0.33$	$3.7 \pm 0.33$	54.02	49.42	44.58
LC <sub>50</sub>	- 1	$8.0 \pm 0.00$	$7.3 \pm 0.17$	$3.3 \pm 0.33$	$3.0 \pm 0.58$	$3.0 \pm 0.00$	36.67	37.75	41.09
4 <sup>th</sup>									10.77
Control	$11.7 \pm 0.33$	$10.7 \pm 0.33$	$11.3 \pm 0.88$	$5.3 \pm 0.33$	$5.3 \pm 0.33$	$5.5 \pm 0.15$	49.53	49.53	48.07
LCm	+1	$9.7 \pm 0.33$	$9.3 \pm 0.33$	$5.0 \pm 0.00$	$4.5 \pm 0.35$	$4.0 \pm 0.58$	51.55	46.39	43.01
LCs		$7.7 \pm 0.33$	$7.3 \pm 0.33$	$4.0 \pm 0.00$	$3.7 \pm 0.33$	#	50.00	48.05	45.20
1 SD between con	11	- 1	$4^{th} = 2.3447$	$2^{nd} = 1.2831$		11			
between time	11		$4^{th} = N. S.$	$2^{nd} = N.S.$		11			
Interaction	11		$4^{th} = N. S.$	$2^{nd} = N. S.$		4 <sup>th</sup> = N.S.			

	$\mathrm{LC}_{9}$	LC 20	Control		Treatment
				72 h.	2 <sup>nd</sup>
				96 h.	2 <sup>nd</sup> larval instar after
0.03				120 h.	ter
03 mm				72 h.	<b>4</b> <sup>th</sup>
		B		96 h.	4 <sup>th</sup> larval instar after
				120 h.	îter

Fig. (20): Cystocyte cell types recognized in untreated and Dipel 2X treated S. littoralis larvae.

## 5.3.2. Quantitative analysis of haemocytes:

# 5.3.2.a. Effect of Dipel 2X on percentages of different haemocytes of the 2<sup>nd</sup> instar larvae of S. littoralis :

Data presented in Table ( 49 ) and Fig . ( 21 ) indicated that the prohaemocytes occupied  $51.29\pm1.25$  ( 49.00-58.29 % ) in the haemolymph of the  $2^{nd}$  instar **S.** littoralis larvae of the total count of haemocytes . Larval feeding on leaves treated with Dipel 2X at LC  $_{20}$  and LC  $_{50}$  levels decreased this average ot 43.32 and 34.61 % , indicating 15.54 and 32.52 % reductions in the haemocyte counts due to these treatments , respectively than control .

As for the phagocyte cells (plasmatocytes, spindle and granular cells ) , those occupied  $20.10\pm0.66$ ,  $4.33\pm0.58$  and  $15.31\pm0.31$ % of the total haemocytes of untreated larva , respectively . While due to treatments by LC<sub>20</sub> & LC<sub>50</sub> , their numbers were found to increase by 32.4 & 63 % ( 26.6 & 32.8 %) , 24.4 & 31.4 % ( 5.37 & 5.96 %) and 6.79 & 28.6 % ( 16.35 & 19.7 %) for the three phagocytic cell types , respectively ( Table , 48 & Fig. 21 & 22 ) .

Slight increase occurred in the adipohaemocytes to occupy 4.00 and 4.12 % of the total number of haemocytes in the  $2^{nd}$  instar larvae treated with Dipel 2X at LC  $_{20}$  and LC $_{50}$ , respectively, opposed to 3.09 in the control larvae . This result indicated increases in the amount of adipohaemocytes in the treated larvae by 29.45 and 33.3 % for LC  $_{20}$  and LC  $_{50}$  levels, respectively.

Oenocytoids , spherule cell and cystocytes in the untreated larvae occupied 1.36 , 2.41 and 2.11 % of the total number of haemocytes , respectively . Treatment with Dipel 2X led to decreases in their cell numbers to occupy 1.21 , 2.12 & 1.02 at LC<sub>20</sub> level and 0.77 , 1.58 a& 0.77 at LC<sub>50</sub> level , respectively . The reduction percentages of the

**Table (49)**: Effect of (Dipel 2X) on percentages of different haemocyte numbers of  $2^{nd}$  instar S. *littoralis* larvae after 5 days of treatment.

Type of haemocyte	Means	± S. E. % an	d range	% reducti increase con	(+) from
	control	LC <sub>20</sub> (0.3300 X 10 <sup>3</sup> )	LC <sub>50</sub> (1.0592X10 <sup>3</sup> )	LC <sub>20</sub>	LC 50
Prohaemocytes	$51.29 \pm 1.25$ (49.00 - 58.29)	$43.32 \pm 0.69$ (40.00 - 46.64)	$34.61 \pm 0.58$ (33.61 - 35.61)	- 15.54	- 32.52
Plasmatocytes	$20.10 \pm 0.66$ $(19.00 - 21.10)$	$26.61 \pm 1.03$ (24.46 – 27.00)	$32.77 \pm 0.44$ (30.00 - 45.54)	+ 32.39	+ 63:03
Spindle cells	$4.33 \pm 0.58$ $(3.33 - 5.33)$	$5.37 \pm 0.29$ $(3.37 - 7.00)$	$5.96 \pm 0.39$ (5.00 - 8.38)	+ 24.40	+ 31:41
Granulated cells	$15.31 \pm 0.31$ (15.00 - 16.93)	$16.35 \pm 0.58$ $(15.35 - 17.35)$	$19.69 \pm 0.75$ $(19.00 - 21.38)$	+ 6.79	+ 28,61
Adipohaemocytes	$3.09 \pm 0.97$ $(2.00 - 4.27)$	$4.00 \pm 0.25$ (3.00 - 5.86)	$4.12 \pm 0.83$ $(3.30 - 6.00)$	+ 29.45	+ 33.30
Oenocytoid cells	$1.36 \pm 0.21$ $(1.00 - 1.72)$	$1.21 \pm 0.07$ $(1.00 - 1.42)$	$0.77 \pm 0.13$ $(0.51 - 0.90)$	- 17.65	- 43,38
Spherule cells	$2.41 \pm 0.50$ $(1.23 - 2.77)$	$2.12 \pm 0.36$ (1.24 - 3.00)	$1.58 \pm 0.33$ $(1.00 - 2.16)$	- 12.03	- 34,44
Cystocyte cells	$2.11 \pm 0.11$ $(2.00 - 2.33)$	$1.02 \pm 0.01$ $(1.00 - 1.04)$	$0.77 \pm 0.06$ $(0.70 - 0.90)$	- 51.66	- 63.51

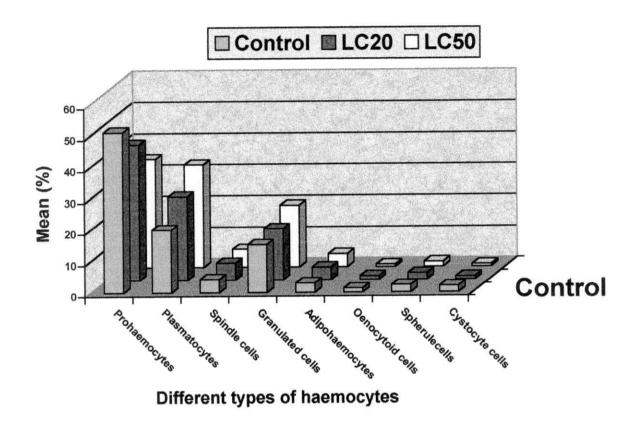


Fig. (21): (DHCs) in  $2^{nd}$  instar S. littoralis larvae after treatment with LC<sub>20</sub> and LC<sub>50</sub> of Diple 2X.

three haemocyte types were 17.65 , 12.03 & 51.66 % at LC $_{20}$  level and 43.38 , 34.44 & 63.51 % at LC $_{50}$  level of Dipel 2X treatment , respectively , ( Table 49 & Figs . 21 , 22 ) .

# 5.3.2.b. Effect on haemocyte counts in 4<sup>th</sup> instar S . littoralis larvae after 5 days of treatment :

When S. *littoralis* larvae were fed on leaves treated with LC  $_{20}$  and LC  $_{50}$  of Dipel 2X , the percentages of prohaemocyte counts in the  $4^{th}$  instar larvae occupied 44.00 and 33.13 % , respectively indicating 14.21 and 35.41 % reduction in number than the 51.29 % estimated in the control larvae (Table 50 and Fig . 22).

In the healthy  $4^{th}$  instar larvae , the phagocytic cells (plasmatocytes , spindle cells and granular cells) occupied 20.1 , 4.33 and 15.13 % of the total haemocyte count , respectively (Table , 50). By treatment with Dipel 2X , those occupied were 25.41 , 5.45 & 17.39 % at LC<sub>20</sub> level and 31.33 , 6.83 & 21.48 % at LC<sub>50</sub> level , respectively . Thus indicating increases by 26.42 , 25.87 & 13.58 at LC<sub>20</sub> level and 55.87 , 57.74 & 40.30 at LC<sub>50</sub> level in the three cell type counts , respectively (Table , 50 and Fig 22) .

The percentage of adipohaemocytes were found to increase also by Dipel 2X treatment to occupy 3.81 and 4.03 % by  $LC_{20}$  and  $LC_{50}$  treatments , respectively , showing 23.30 and 30.42 % increase than the 3.09 % estimated in control larvae (Table , 50).

Concerning the remaining types ( Oenocytoids , spherule cell and cystocyted ) , all decreased in percentages due to bioinsecticide treatment as those they occupied 1.00 , 1.44 & 1.50 % at LC<sub>20</sub> , respectively , and 0.80 , 1.4 and 1 % at LC<sub>50</sub> level , respectively opposed to 1.36 , 2.41 and 2.11 % in the untreated larvae , respectively . These data indicated

Table ( 50 ): Effect of (Dipel 2X) on percentages of different haemocyte numbers of  $4^{th}$  instar S. *littoralis* larvae after 5 days of treatment

Type of haemocyte	Means	± S. E. % an	d range	% reducti increase con	** · ·
	control	LC <sub>20</sub> (0.4900 X 10 <sup>3</sup> )	LC <sub>50</sub> (1.8432X10 <sup>3</sup> )	LC <sub>20</sub>	LC <sub>50</sub>
Prohaemocytes	$51.29 \pm 1.25$ (49.00 - 58.29)	$44.00 \pm 0.59$ (43.00 - 45.00)	$33.13 \pm 1.55$ $(30.00 - 35.26)$	- 14.21	- 35.41
Plasmatocytes	$20.10 \pm 0.66$ $(19.00 - 21.10)$	$25.41 \pm 0.20$ $(25.00 - 25.82)$	$31.33 \pm 1.07$ (26.00 – 35.00)	+ 26.42	+ 55.87
Spindle cells	$4.33 \pm 0.58$ $(3.33 - 5.33)$	$5.45 \pm 0.84$ $(3.37 - 7.00)$	$6.83 \pm 0.48$ (5.00 - 7.66)	+ 25.87	+ 57.74
Granulated cells	$15.31 \pm 0.31$ (15.00 - 16.93)	$17.39 \pm 0.22$ (17.00 - 18.78)	$21.48 \pm 0.85$ (19.00 - 21.96)	+ 13.58	+ 40.30
Adipohaemocytes	$3.09 \pm 0.97$ $(2.00 - 4.27)$	$3.81 \pm 0.85$ $(3.00 - 5.96)$	$4.03 \pm 0.41$ (4.03 - 7.03)	+ 23.30	+ 30.42
Oenocytoid cells	$1.36 \pm 0.21$ $(1.00 - 1.72)$	$1.00 \pm 0.06$ (1.90 - 1.10)	$0.80 \pm 0.17$ $(0.50 - 1.10)$	- 26.47	- 14.18
Spherule cells	$2.41 \pm 0.50$ $(1.23 - 2.77)$	$1.44 \pm 0.25$ $(1.00 - 1.88)$	$1.40 \pm 0.57$ (0.90 - 1.90)	- 40.25	- 41.91
Cystocyte cells	$2.11 \pm 0.11$ $(2.00 - 2.33)$	$1.50 \pm 0.29$ $(1.00 - 2.00)$	$1.00 \pm 0.15$ $(0.80 - 1.30)$	- 26.07	- 52.61

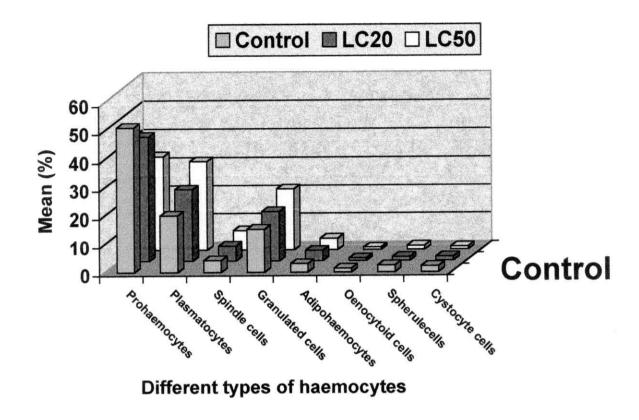


Fig. (22): (DHCs) in  $4^{th}$  instar S. littoralis larvae after treatment with  $LC_{20}$  and  $LC_{50}$  of Diple 2X.

that larval treatment with Dipel 2X led to reductions in the cell percentages by 26.47 , 40.25 & 26.07 at LC $_{20}$  level and 14.18 , 41.91 and 52.61 % at LC $_{50}$  level , respectively , ( Table , 50 ) .

From the previously explained results it could be concluded that the prohaemocytes occupied, generally, most of the haemocyte counts in  $\bf S$ . *littoralis* larvae (more than 50% of the total haemocyte counts). Plasmatocytes came the next 20.10 % of the total haemocyte counts, that was followed by the granulated cells 15.31 and spindle cells 4.33 %. Each of the remaining 4 haemocyte types, adipohaemocytes, oenocytes, spherule cells and cestocytes ocupied only 1.36-3.1% of the total haemocyte count.

The obtained data indicated also that larval feeding on leaves treated with Dipel 2X at  $LC_{20}$  and  $LC_{50}$  reduced the numbers of prohaemocytes , oenocytoids , spherule cells and cystocytes than those estimated in the haemolymph of the untreated larvae .While , on the contrary , the percentages of phagocytes ( Plasmatocytes , spindle cells and granulated cells ) increased in the treated larvae than control . This appears normal due to the active defense mechanism of phagocytes against any strange materials (  $\emph{B.t. kurstaki}$  in this investigation ) that came into the insect haemolymph .

In accordance to the present results, *Miselyunene* (1976) noticed that the numbers of proleucocytes, oenocytoids and micronucleuctes in the haemolymph of the cabbage butterfly larvae treated with endobacterin were reduced with the development of infection. *Abdel* – *Rahim et al* (1994) reported reduction in the total haemocyte counts (THC s) in the haemolynph of *Artogeia rapae* after treatment of 4<sup>th</sup> instar larvae by Bactospien at different concentrations. *Kares et al* (1992)

reported decreases in the prohaemocytes and increases in the plasmatocyted and spindle & granular cells due to treatment of A. rapae by Bactospeine at the  $LC_{10}$  and  $LC_{50}$ . Also, Kares (1994) indicated the parasitism by Zele nigricornis in the  $2^{nd}$  instar larvae of S. Ittoralis resulted in decrease in the prohaemocyte count and an increase in tha phagocytic cells ( plasmatocytes , spindle and granular cells ). The same result was also indicated by Abd El - Hameed (1995) due to treatment of freshly hatched P. gossypiella larvae with Delfin ( B. thuringiensis kurstaki) at the  $LC_{20}$  and  $LC_{50}$  levels .

But in contrast to the presen results, *Chain* and *Anderson* (1983) reported that injection of suspension of B. cereus into the haemocol of G. mellonella larvae caused a rapid decrease in the number of circulating plasmatocytes.

# 6 . Histopathological effect of Dipel 2X and MVP $\Pi$ on larval of ${\it S.littoralis}$ :

### 6.1. Histology of normal larvae:

The midgut of a normal larva (Fig. 23, A) consists of a single cellular layer resting upon a basement membrane, which is surrounded externally first by circular, then longitudinal muscles. The cellular layer (the epithelium) consists of columnar cells, each with a large granular nucleus. The epithelial cells are interspersed with cluster of small regenerative cells, also resting upon the basement membrane. Each regenerative cell contains a relatively large nucleus and strongly basophilic cytoplasm. The lumen is surrounded by a peritrophic membrane.

# 6-2- Histological effect of Dipel 2X on larvae of S. littoralis:

The microscopic examination of treated and untreated larval midgut section (Figs., 23 to 26) indicate that there were differences between midgut sections of treated and untreated larvae.

### 1<sup>st</sup> larval instar treatment :

The histopathological effects caused by LC<sub>50</sub> and LC<sub>90</sub> of Dipel 2X on the midgut of  $1^{st}$  instar larvae involved separation of epithelial cells from the basement membrane, and in some cases the basement membrane (Figs. 23 F and 24 E), and slight vacuolation appeared in some cases. The epithelial cells of the midgut in most areas showed degeneration (Figs. 23 D, E & F and 24 C & F). In some cases the epithelial cells appeared deformed (Figs. 23 C and 24 D). Also,

Dipel 2X caused disorganization and disintegration of peritrophic membrane ( Figs. 23 C , D , E & F and 24 B ,

## ♦ 3<sup>rd</sup> larval instar:

Strong histopathological symptoms were detected in the midgut of larvae fed on castor — bean leaves treated with Dipel 2X at LC50 and LC90 . The epithelial cells of the midgut in most areas showed marked degeneration ( Figs . 25 B , C , E & F and 26 A , B , D & F ) . It is evident that scattered groups of the epithelial cells were sloughed off into the lumen . ( Figs . 25 A , B & F ) . Cell basement disappeared ( Figs.25 B , E & F and 25 F ) and slight vacuolation appeared in most cases . In some cases , the epithelial cells appeared deformed and became of smaller size than those of the control larvae ( Figs. 25 D & G and 26 C ) .

Damaging the gut epithlium of *Anagasta kuhniella* larvae after ingestion of *B. thuringiensis* spores was previously reported by *Mattes (1927)*. *Tonada (1953)* described disorganisation of the midgut epithelium of *Pieris rapae* infected by *B. thuringiensis*. As the infection advanced, the destraction of the mid-gut wall continued until most of the epithelium was destroyed. Also, *Kinsinger* and *Megauchey (1979)* indicated that infection of *Plodia interpunctella* and *Ephestia cautella* larvae by *B. thuringiensis* supsb. *Kurstaki* spores and parasporal crystals caused degeneration of mid-gut epithelial cells, progressively, until the mid-gut was totally disrupted. *Hamed (1979)* studies the etiological effect of *B. thuringiensis* on *S. littoralis*. The author found that infection caused the epithelial cells to be disorganized at detached from the basement membrane 24 & 48 hours after treatment *Salama et al (1993)* indicated that *S. littoralis* larvae treatment with *B. thuringiensis* caused cell hypertrophy with

some deposits of B.t. crystals on the  $4^{th}$  day after infection. The authors concluded that the resultant initial destruction caused by the B.t. toxin facilitated penetration of vegetative cells and spores of B.t. in the mid-gut epithelial cells.

### 6 -3 - Histological effect of MVP $\Pi$ on larvae of S. littoralis :

The MVPII preparation induced rapid effect on the mid-gut epithelium. After feeding, the toxins had bound to the peritrophic membrane (Figs. 27 E & F, 28 C, E & F, 29 A, E & F and 30 C, D & E). Some damage to the epithelium. such as disruption of the cell and vacuolization of the cytoplasm, was already apparent, (Figs. 27 D, E & F, 28 B, D, E & E, 29 A, B & F and 30 A & F). According to *Yi et al (1996)* it is thought that forced feeding of *S*. *littoralis* larvae led to leakage of the cytoplasmic contents from the damaged cells into the gut lumen, and binding of the toxins to the peritrophic membrane (Figs. 29 B, E & F). In some cases, the epithelial cells appeared deformed (Fig. 29 A & F), and became of smaller in size than control Fig. 30A & E).

Working of the effect of  $\delta$  endotoxin, *Gerald and Earle* (1966) indicated that feeding of *Ostrinia nubilalis* larvae on cystalline paraspores of *B. thuringiensis* var. *thuringiensis* caused the mid-gut epithelial cells to slough off the lumen and thus expanse areas of the basement membrane became exposed to attack by the vegetative bacteria and the crystal.

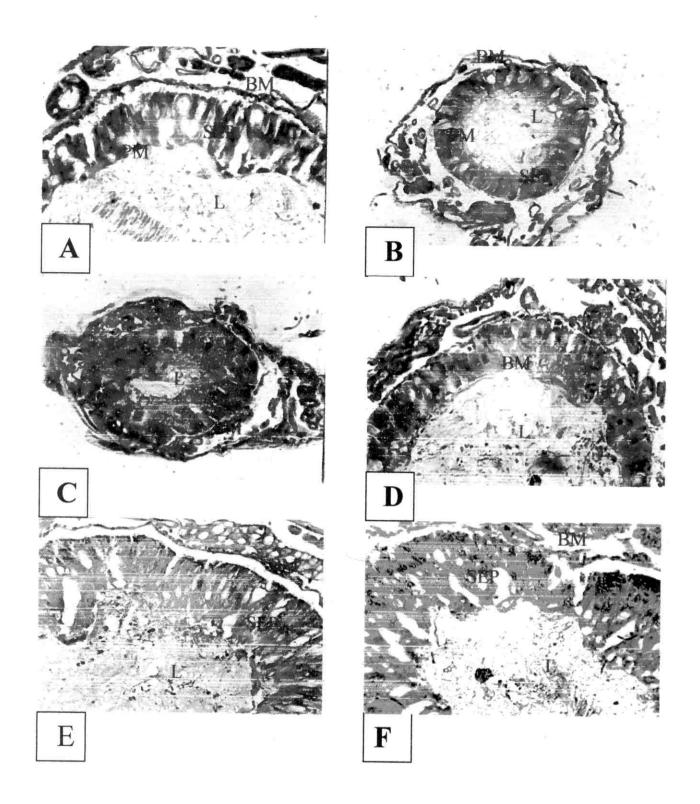


Fig. (23): Transverse sections in the midgut of the  $1^{st}$  instar S. littoralis larvae before treated (A) and after 24h. (B), 48h. (C), 72h. (D), 96h. (E) and 120h. (F) of castor – bean leaves treated with LC<sub>50</sub> of Dipel 2X (100X).

BM = basement membrane PM = peritrophic membrane

SEP = epithelial cells

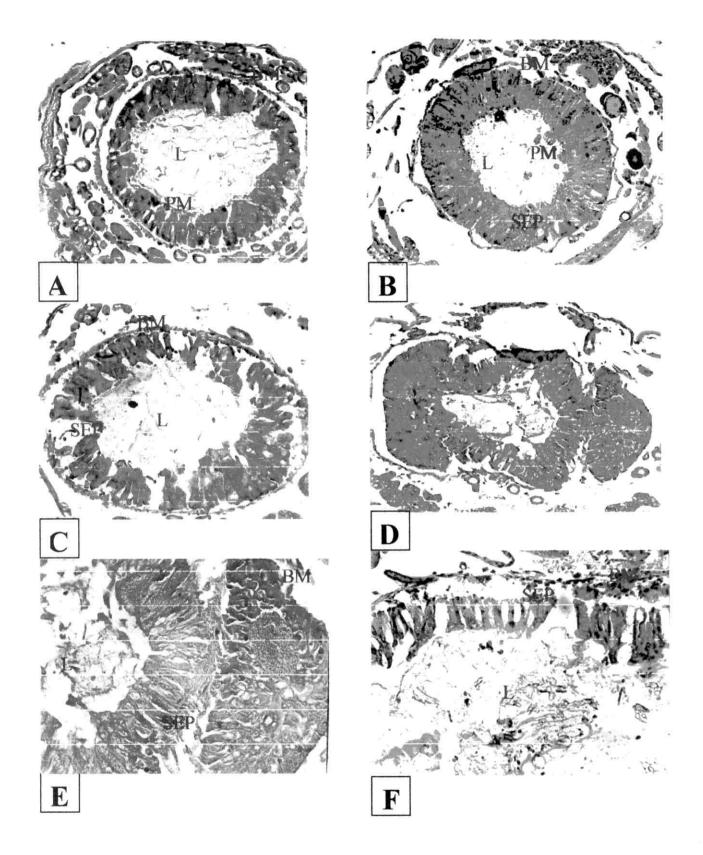


Fig. (24): Transverse sections in the midgut of S. littoralis  $1^{st}$  instar larvae after 24h. (A), 48h. (B), 72h. (C), 96h. (D&E) and 120h. (F) of feeding on castor – bean leaves treated with LC<sub>90</sub> of Dipel  $2X (100 \ X)$ .

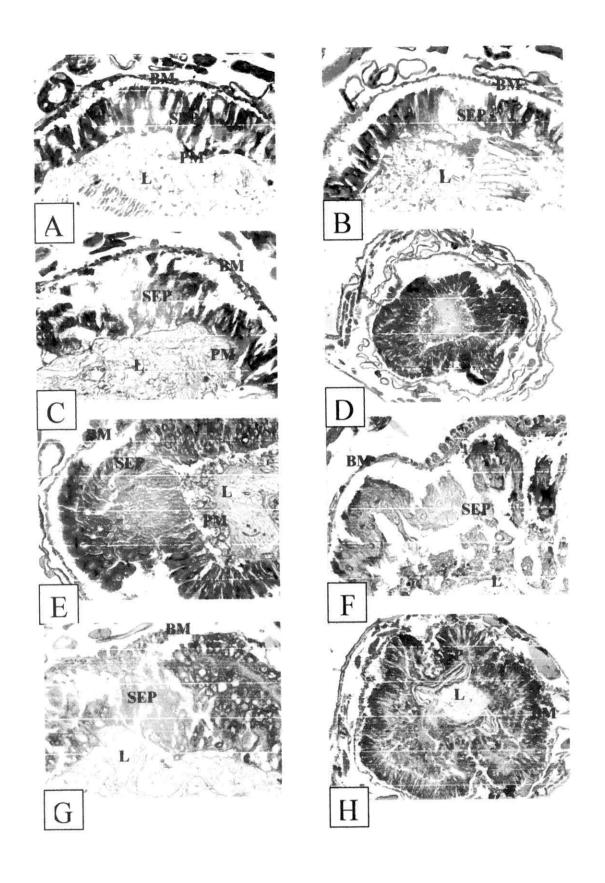


Fig. (25): Transverse section in the midgut of the control 3<sup>rd</sup> instar S. littoralis (A), 24h. (B), 48h.(C), 72h..(D&E) and 120h.. (G) feeding on leaves treated with LC<sub>50</sub> of Dipel 2X (100X).

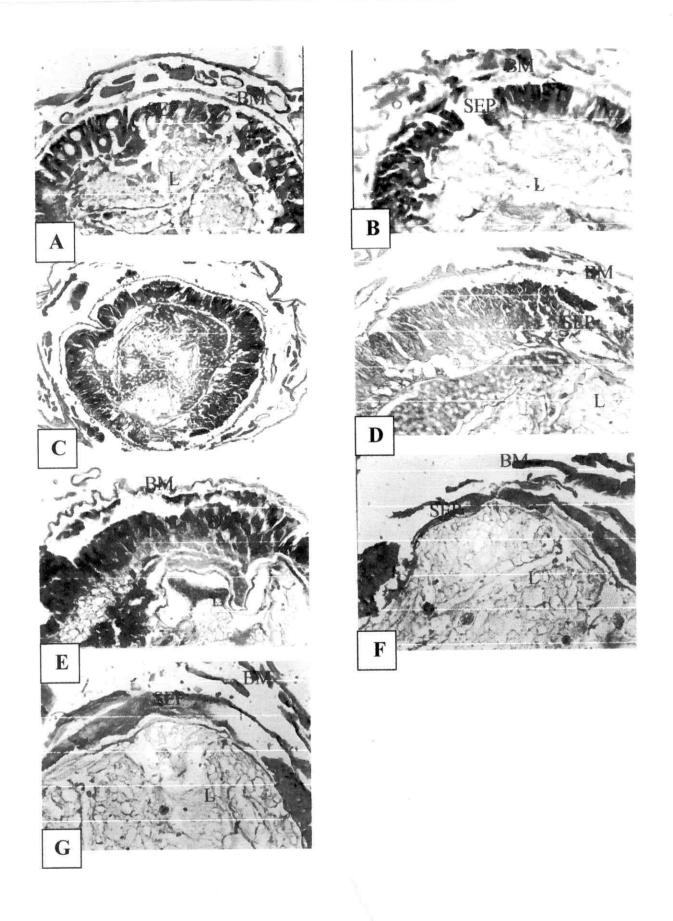


Fig. (26): Transverse sections in the midgut of the 3<sup>rd</sup> instar S. littoralis larvae after 24h.(A), 48h. (B), 72h. (C)&D), 96h. (E) and 120h. (F&G) of feeding on leaves treated with Dipel 2X (100X).

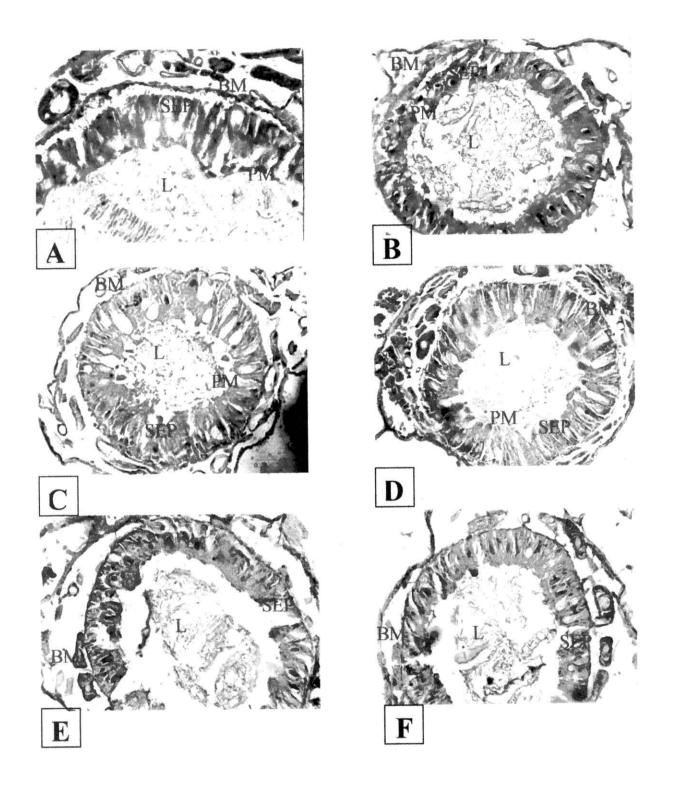


Fig. (27): Transverse sections in the midgut of untreated S. littoralis  $1^{st}$  instar larvae (A) and after 24h. (B), 48h. (C), 72h. (D), 96h. (E) and 120h. (F) of feeding on castor – bean leaves treated with the LC<sub>50</sub> of the MVP  $\Pi$  (100 X).

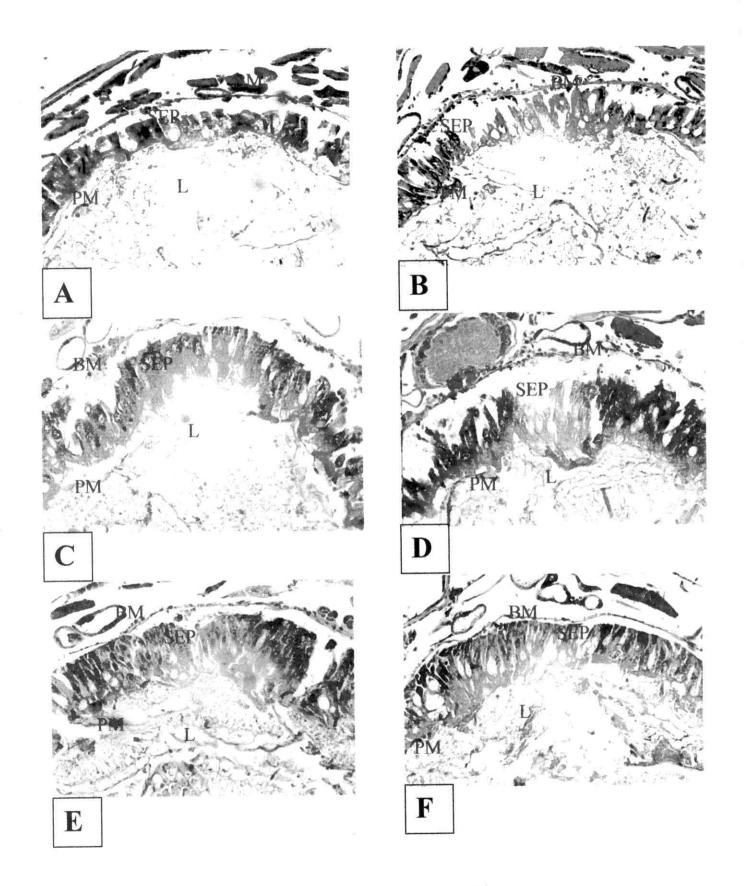


Fig. (28): Transverse sections in the midgut of the 1<sup>st</sup> instar S. littoralis larvae after 24 h. (A), 48h. (B), 72h. (C), 96h. (D & E) and 120h. (F) of feeding on castor – bean leaves treated with LC90 of MVP Π (100X).

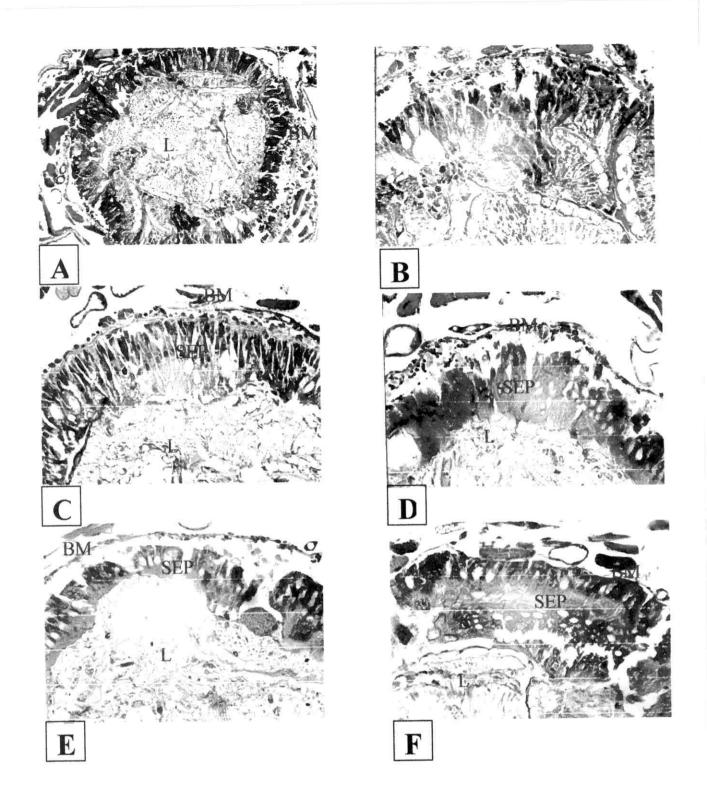


Fig. (29): Transverse sections in the midgut of S. littoralis  $3^{rd}$  instar after 24h. (A&B), 48h. (C), 72h. (D), 96h. (E) and 120 h. (F) of feeding on castor – bean leaves treated with LC  $_{50}$  of MVP $\Pi$  (100 X).

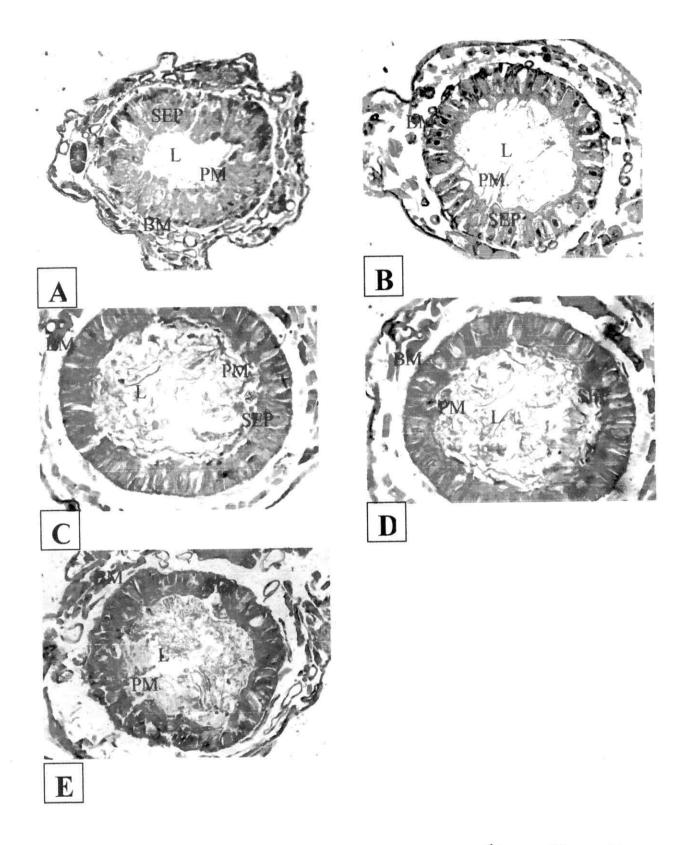


Fig. (30): Transverse sections in the midgut of the  $3^{rd}$  instar **S.littoralis** larvae after 24h. (A), 48 h. (B), 72h. (C), 96h. (D) and 120 h. . (E) of feeding on castor – bean leaves treated with LC<sub>90</sub> of MVP  $\Pi$  (100 X).