Toxicological and Biochemical Effects of Some Folair Nutrients and Alternative Compounds on The Spiny Bollworm, *Earias Insulana*(Boisde.) (Lepidoptera: Noctuidae)., In Relation to Larval Electrophoresis.

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ABSTRACT

The present study was carried out to evaluate the efficiency of the insecticide- Baythroid®, six nutritive foliars , and a phytochemical compound; tested in a trial of initiating an Integrated Pest Management programme (IPM) for the spiny bollworm *(Earias insulana)* either alone or in consequence upon the biofertilized cotton plants of "Giza-70" during the two successive seasons of 2004 and 2005 .The side effects of the above mentioned materials were also studied on cotton yield. The obtained results declared that the applied tri and /or bi - sequent sprays of Baythroid® with Greenzit N.P.K and Neem oil or with Greenzit $S.P_{100}$ in the season 2004, efficiently decreased the spiny bollworm infestation to the level of 0.81and 0.87 larvae/10 bolls, in respect. The treatment of Baythroid®with potasin-f and Greenzit N.P.K was less efficient 1.54 larvae/10 bolls. The performed tri-consequent sprayings of Baythroid®with Greenzit $S.P_{100}$ and Ascorobic acid; Baythroid® alone and/ or with polymex ; Ascorbic acid were efficient and resulted in high reduction of the infestation level. The made tri- sequent sprayings of Baythroid®with polymex and potasin-f and/ or with potasin-f and Greenzit N.P.K, which gave a relatively higher level of the spiny bollworm infestation.

Moreover, the bi-sequent and/or tri-sequent sprayings of Baythroid® with Greenzit $S.P_{100}$; Baythroid® with Greenzit N.P.K and Neem oil on the bio-fertilized plants, similarly Baythroid® with Greenzit $S.P_{100}$; Baythroid®with Greenzit $S.P_{100}$ and Ascorobic acid indicated the highest cotton yield in the seasons of 2004 and 2005, respectively. Vice versa, the treatment of Baythroid® alone or in sequence with potasin-f and Greenzit N.P.K showed the lowest cotton yield. The total protein analysis on the sampled spiny bollworms larvae *(Earias insulana)* from each of the eight performed treatments indicated obvious variability in the protein pattern of the different inspected larvae using sodium dodecyl sulphate (SDS-PAGE) polyacrylamide gel electrophoresis.

INTRODUCTION

The cotton bollworms are regarded as one of the most destructive insect-pests of cotton plants in Egypt . Cotton is considered the preferred host plant for bollworms.

The indiscriminate use of insecticides has caused a number of ecological, economical and social problems worldwide including Egypt, besides the proved resistance of several insect pests to chemicals.

Foliar fertilization using different formulated nutritive elements might be a good tool to produce a tolerant and profitable cotton plants that compete with weeds and able to outgrow and overcome diseases and insect damage (Mesbah *et.al.* 2000,2004 and 2005). Foliar nutrients can also correct the resulting deficiencies due to the lack of certain nutrients, which are required in large amounts (macro-elements) and / or required in trace amounts (micro - elements) (Locke and Eck, 1965 ,Michaeal Treshow, 1970 and El-Naggar,1998).

In addition, biochemical assay may be a better choice for detecting resistance at lower gene frequencies than bioassays (Brown and Brogdon 1987) .The most accurate way to monitor the pink bollworm susceptibility is through the bioassay and/ or the biochemical techniques (Devonshire *et al* .1986). Electrophoretic techniques permit precise and relatively simple examination of genetic differences among insect individuals and among insects populations. (Abdel- Sarnie *et al* 1979 Abdel – Hafez *et al* 1983 and Abdel - Hafez 1985).

However, some differences between treated larvae and field collected larvae were noticed in the protein molecular weight. Similar differences were found between susceptible and feral larvae and also susceptible and wild type moths (Shekeban 2000).

Therefore, this study was directed to: (i) evaluate certain treatments of some macro, micro foliar nutrients and alternative chemical compounds that have been tested either alone or in bi and/ or tri consequent applications, as a pest control measure against the spiny bollworm, (ii) study the the above mentioned materials on cotton yield and (iii) determine any possible changes in the protein patterns of the treated larvae.

MATERIALS AND METHODS

Field experiments were carried out at the experimental farm of the Faculty of Agriculture, Saba Basha, Alexandria University, Egypt, during the two successive growing cotton seasons of 2004 and 2005. In both seasons, an area of half Feddan was cultivated with cotton cv. "Giza70" on April the 8th, 23rd and 18th, respectively. The used experimental design was the split design with three replicates as well as untreated check.

Prior planting in both seasons the cotton seeds were treated with the bio-fertilizer "Microbin®" and then sown at 25 cm apart between hills with spacing of 60 cm between furrows. The experimental area consists of 8 treatments, in

addition to an untreated check. Treatments including foliar fertilizers, alternative chemical compounds and the insecticide Baythroid® with different chemical combinations were evaluated. Rates of the tested compounds are shown in Table (1).

The applications of each tested compound were applied at the beginning of flowering period, period of fifty percentage of flowering, the beginning of fruiting throughout the whole season. Weekly inspections were performed to determine the efficiency of the tested compounds on the spiny bollworm *(Earias insulana)* larvae. Sprayings were done using knapsach sprayer (20L).

I - Estimation of the spiny bollworm infestation:

Prior to the application of foliar treatments, the primary infestation level of the spiny bollworm was determined later. Weekly samples of 10 green bolls out of 30 green bolls / treatment were taken randomly, to estimate the level of infestation during seven weeks in each season.

II -Determination of cotton yield:

In each treatment ripened open bolls from twenty- five cotton plants were collected to determine the rate of cotton yield/plant, from which, the total yield/feddan was mathematically calculated.

III- Statistical analysis:

Using "F" and "L.S.D" tests were performed for detecting the difference and significancy of recorded results. The % increase of each yield over the check was calculated according to the formula of H. Nagwa as follows:

% increase = (A-B) / B * 100

A = Cotton yield of each treatment

B = Cotton yield of check

IV- The tested foliar fertilizers and chemical compounds:

1. Foliar fertilizers:

a- Greenzit S.P 100:

The main components are:

EDTA	Na_2 MN	40%
EDTA	Na_2 Z	43%
Fe,Mg,Mn,B,Zn,Cu,Mo,Ni,Co		12%

Concentrations of Ferric and micro-elements in mg/kg:

Co	0-054	Fe	5.40	Mo	0.027	Mg	0.54
Ni	0.005	Mn	5.54	Cu	0.005	Zn	70.27

b- Greenzit N.P.K. 5144:

The main compounds are:

Micro-elements mg/L		Macro-elements gm/L	
Fe	1000	N	70
Mg	100	P	30

Mn	100	K	39
B	100		
Zn	50		
Cu	10		
Mo	5		
Ni	1		
Co	1		

c- Potasin F:

The main components are: N.P.K. (0:10:30), 30% potassium oxide, 10% fifth potassium oxide.

d- Polymex:

Containing Zn, Fe, Mn, cu, B, Mg, (supplied by Mesbah in 2001) was used.

e- Ascorbic acid:

($C_6H_8O_6$) white to slightly yellow crystals was included in some treatments.

4-2-Cyfluthrin: (Baythroid®):

It is a synthetic Pyrethroid used in the form of E.C. 50% formulation for insect control.

It's chemical name is : (RS) -ce - Cyano- 4- fluro-3 ce Phenoxbenzyl (1 Rs, 3Rs= 1 Rs,3SR)-3- (2.2-dichlorovinyl) -2.2-dimethlcycloprop - anecarboxylato.

4-3-Neem Oil:

In the form of Azadriac ıtin Emulsifiable concentrate (1500 ppm.).

V - Sample preparation :

Ten larvae from each strain were weighed, mass homogenized in 10 ml of 0.02 M sodium phosphate buffer(pH 7.0) using a glass homogenizer with a Teflon pestle. The crude extract was centrifuged at 5000 rpm for 20 minutes using a cooling centrifuge, then passed through glass wool to remove the remaining insoluble cell debris and lipids. The supernatant served as the homogenate and used for the performed assays.

Total protein was extracted by the method of Bradford (1976) and the samples were diluted to 1mg protein/1 ml, and frozen immediately at -4°C until analysis.

VI - Protein electrophoretic analysis:

The performed separation of the general protein patterns in larval homogenates was carried out on slab gels that contained 10% acrylamide and 0.8% bisacrylamide in the presence of sodium dodecyl sulphate (SDS) under reducing conditions, by using the modified method of Laemmli (1970).

The gel was electrophorezed at –4°C at 75 mA / gel until tracking dye (Bromophenol blue Sigma St. Louis, Mo. USA) was injected within 1 cm of the upper gel margin. Gel was stained using 0.12 %W/V Comassie brilliant blue (R-250, Sigma). Seven molecular weight standards were used as markers. Phoretic ID image analysis system (Phoretix International, Lodon) was used to integrate the data of the protein bands.

The molecular weights of the bands on each-gel were then scanned for electrophoretic analysis. Results were graphically depicted with Image Phoretic (IAD) Quantifier Software (Phoretix International,London). The molecular weights were used for a relative comparison of each band on the same gel.

RESULT AND DISCUSSION

1- Comparative efficiency of the tested insecticide Baythroid® and the nutritive foliars on the spiny bollworm infestation:

a. Season of 2004:

Included results in Table (2) showed the calculated mean numbers of the inspected spiny bollworm larvae per ten bolls in each of the conducted treatments for the determination of the effect of tested chemicals on the infestation incidence of the assigned insect-pest.

From table (2) it could be noticed that each of the performed bi-and/or tri-sequent sprays of Baythroid®with Greenzit $S.P_{100}$ or with Greenzit N.P.K and Neem oil decreased efficiently the spiny bollworm infestation to 0.81 and 0.87 larvae/10 bolls, successively. On the contrary, the other applied foliar treatments, with rate of infestation more or less increased from 0.96 to 1.30 larvae/10 bolls. Except for, the treatments of Baythroid®with potasin-f and Greenzit N.P.K and / or the untreated check, indicated that the level of infestation increased up to 1.54&1.42 larvae/10 bolls, in respect.

b- Season 2005:

The exhibited data in Table (3) showed the detected response of the bio-fertilized cotton plants to the performed foliar treatments. The revealed effects of each foliar treatment were expressed in mean numbers of *Earias insulana* larvae/10 bolls. It was concluded also that in comparison to the untreated check (1.63 larvae /10 bolls) the performed tri-sequent sprays of Baythroid®/ Greenzit $S.P_{100}$ / Ascorbic acid followed by Baythroid®alone gave a highly efficient control of the spiny bollworm (0.63&0.87 larvae/10 bolls, respectively). That lower level of spiny bollworm infestation slightly increased representing a range of 0.90-1.18 larvae/10 bolls compared with the most of the other applied foliar treatments that increased up to 1.27 larvae/10 bolls in case of the tri-sequent sprays of Baythroid®/ polymex /

potasin-F.

Obtained results of the present work are in agreement with Mahasen M. Ibrahim *et. al* (2000) who confirmed that the highest efficiency in reducing the percentage of infestation with both the pink and the spiny bollworms was recorded for the treatment application of Baythroid® [Cyfluthrin] , which gave reduction rates of 91.55%, followed by Fenvalerate (88.43%), ES, Fenvalerate (85.33%) and Fenpropathrin (83.64%) in a descending order.

Moreover, Mahasen Abdel- Aziz (2002) showed that the foliar application of Potassium foliar fertilizer (38% Potassium)and Potasin- F (30% Potassium oxide and 10% Phosphor dioxide) at a rate of one liter/fed., as well as spraying with Ascorbic acid (30 g/fed.) and / or Salicylic acid (30 g / fed.), in sequence, including greenhouse and field trials had a significant effect on reducing percentages of the spiny bollworm infestation and increasing actual yield in comparison to untreated plots.

Mesbah *et al* (2004) declared the superior effect of Baythroid®with each of Greenzit formulations alone and/or sequented by the other tested foliar chemicals on the pink bollworm infestation, whereas the bi-and/or tri sequent sprays of Baythroid®with Greenzit $S.P_{100}$ or Greenzit $S.P_{100}$ followed by Ascorbic acid lowered the pink bollworm infestation to-0.09 Larva/10 bolls. They also retreated that in all the initiated treatments of mono, bi, tri and/or tetra sequent sprayings of tested chemicals lowered to a great extent not exceeding 0.33-0.04 larva/10 bolls in both seasons, the mean numbers of inspected larvae *(Earias insulana)*.

In addition, Mesbah *et al* (2005) evaluated the efficiency of Baythroid®, nutritive foliars and alternative chemical compounds, which were tested either alone or in consequence upon the bio fertilized cotton plants of "Giza-70" during the three successive seasons (1999 – 2001) in an Integrated Pest Management programme (IPM) of the spiny bollworm *(Earias insulana)*. The side effects of the used compounds on cotton yield losses, crude fat and moisture content of cotton seeds were also studied. The same authors mentioned that the applied bi and /or tri- sequent sprays of Baythroid® with the other tested foliars, more or less decreased efficiently the spiny bollworm infestation to 0.29 and 0.33 larvae/10 bolls. The treatment of Sirene was less efficient (1.95 larvae/10bolls) in controlling the insect pest.

2- Effect of foliar applications on cotton yield:

Results of the conducted field experiments throughout the growing cotton season of 2004 indicated that the performance of bi-sequent sprays of Baythroid®with Greenzits $S.P_{100}$ and/or tri-sequent sprays of Baythroid®with Greenzit N.P.K and neem oil on the bio - fertilized cotton plants with

"Microbin®" registered the highest values of cotton yield (6.2&6.09 kent/fed.).

Vice versa, the consequent spraying of Baythroid® with Greenzit N.P.K and Potasin- F followed by Baythroid® with polymex and Potasin- F gave the lowest values of 4.8 & 5.1 Kent / fed., compared to the untreated check (4.7 kent/fed.) (Table, 4).In season 2005 the carried out tri- sequent sprays of Baythroid®with Greenzit $S.P_{100}$ and Ascorbic acid and/or Baythroid® with Greenzit $S.P_{100}$ produced the highest cotton yield that amounted to 4.5 and 4.4 kent. /fed, in respect. While, the consequent spraying of Baythroid® with Greenzit N.P.K and Potasin- F led to the lowest cotton yield (3.5 Kent. /fed.). But both the treatments of Baythroid® and the untreated check were identical (2.6 kent./ fed.) (Table, 5).

In both seasons of this study, the increase or decrease in cotton yield was due to the differences in the infestation levels. In other words, there was a reversed relationship between the yield and the resulted number of *Earias insulana* larvae owing to the different treatments of this study.

Similarly, Mesbah *et al* (2004) opined that the tri- sequent sprays of Baythroid® with Greenzit N.P.K., Neem oil and/or Greenzit $S.P_{100}$/Ascorbic acid in season 2000 and/or Baythroid®with Greenzit $S.P_{100}$ /Ascorbic acid and/or Potasin F/Greenzit N.P.K. in season 2001 gained higher yields (13.1 & 12.6) and (8.5&8.5kent./fed.) corresponding to lower levels of bollworm infestation of (0.84% and 1.8%) and (0.9 and 1.4%) in the following seasons, in sequence.

Mesbah *et al* (2005) also stated that for the bio-fertilized plants, the tri-sequent sprayings of Baythroid® with Greenzit N.P.K and Neem oil gave the highest value of gained yield .The application of Thyroxine with Greenzit N.P.k and / or Thyroxine with Greenzit $S.P_{100}$ and Ascorbic acid gave higher levels of insect infestation and lowest values of cotton yield in season 2000 and 2001, respectively. In addition, they showed that the grown plants from treated seeds with Thyroxine in season of 2000 and bi-sequently sprayed by Baythroid®with Greenzit N.P.K gained a higher yield, coincided with a lower level of the spiny bollworm infestation, vs., the treatments of potasin-f or Greenzit N.P.K alone and / or potasin-f followed by Greenzit N.P.K and Ascorbic acid, which gave lower of cotton yield as a result of higher percentages of the spiny bollworm infestation.

3- Total Protein Electrophoretic Analysis.

a. Season of 2004:

SDS polyacrylamide gel electrophoresis of the larval protein fractions was assayed in overall samples (control and seven samples,Fig. ,1 and Table,6). A total of 122 amplified fragments occurred overall the samples, with molecular weight ranging from 14.6 to 163 KDa. The control exhibited

the maximum band number (19 bands), while the minimum band number (13 bands) occurred with the Baythroid/Polymex/Potasin-F and Baythroid/Greenzit N.P.K/Neem oil treatments. The total polymorphic band number was 56 bands (45.26%). The lowest number of polymorphic bands (5 out of 13) occurred with the Baythroid®/Polymex/Potasin-F and Baythroid®/Greenzit N.P.K/Neem oil treatments (38.5%), while the highest number (10 out of 19) occurred with the control (52.6%). However, 8 bands were common (monomorphic) for overall control and seven treatments (M.Ws. 163, 70.8, 56.5, 42.7, 37, 31.8, 28 and 25.8, respectively). These results indicated that the most visible changes in protein profile occurred in the Baythroid®/Greenzit $S.P_{100}$/Ascorbic acid treatment where bands with molecular weights of (79.0 and 67.1 k Da) were absent, while they appeared in the other treatments and control. The results also indicated that band with molecular weight (30.0 KDa) is absent in the Baythroid® Polymex/ Potasin-F treatment, but it appeared in other treatments.

Specific markers that generated from SDS-Protein profile were shown in Table (6). Two of 122 protein bands were found to be useful as specific markers. There was one specific band scored with the control and Baythroid®/Greenzit $S.P_{100}$/Ascorbic acid treatment (22.3 and 14.6 KDa, respectively), while there was no specific markers scored with the other treatments. These specific markers could be used in subsequent experiments to detect protein marker for polymorphic genes with economic importance among these and other samples.

b. Season of 2005:

SDS polyacrylamide gel electrophoresis of the larvae protein fractions was assayed in overall samples (control and seven samples, Fig.,2 and Table,7). A total of 147 amplified fragments occurred overall the samples, with molecular weight ranging from 9.0 to 204.3 KDa. The treatment with Baythroid®/Greenzit N.P.K/Neem oil exhibited the maximum bands number (23 bands), while the minimum band number (11 bands) occurred with the Baythroid®/Polymex/Potasin-F. The total polymorphic band number was 115 bands (78.2%). The lowest number of polymorphic bands (7 out of 11) occurred with the Baythroid®/Polymex/Potasin-F (63.6%), while the highest number (19 out of 23) occurred with the Baythroid®/Greenzit N.P.K/Neem oil (82.6%). However, 4 bands were common (monomorphic) for overall control and seven treatments (M.Ws. 69.2, 64.3, 44.2 and 29.7, in respect). Results assured that the most visible changes in protein profile occurred in the Baythroid® /Polymex /Ascorbic acid acid treatment, where bands with molecular weights of 77.6and 35.5 kDa were absent, while they appeared

in the other treatments and control. This result also indicated that band with molecular weight of 70.3 KDa is absent in the Baythroid®/Polymex/Potasin-F treatment, while it appeared in other treatments.

Specific markers generated from SDS-Protein profile were shown in Table (7). One of 147 protein bands were found to be useful as specific marker. There was one specific band scored with the Baythroid®/Potasin-F/Greenzit N.P.K treatment (9.0 KDa), while there was no specific markers scored with the other treatments.These specific marker could be used in subsequent experiments to detect protein marker for polymorphic genes with economic importance among these and other samples.

Poly acrylamide gel electrophoresis is considered as one of the most reliable technique for the detection of changes in cellular protein synthesis, therefore, the protein pattern can be used as a criterion for detecting the differences between several strains and/or individuals exposed to some factors. From the above cited results it could be concluded that the detected presence and/or absence of protein bands in the different adopted foliar treatments in both growing seasons of bio fertilized cotton plants, might be attributed to the occurring changes in the prevailing climatic conditions throughout these growing seasons, which finally reflected on the phenology and physiological status of treated plants and consequently on the biological and physiological status of developing insect-pest larvae within the flowers and/or the growing bolls of these plants.

Identical findings were detailed in the works of many researchers, i.e. Brown and Brogdon(1987)stated that biochemical assay may be a better choice for detecting resistance at lower gene frequencies than bioassays.It was also shown that the most accurate way to monitor pink bollworm susceptibility is through the bioassay and/ or biochemical techniques (Devonshire *et. al.*1986).Abdel-Hafez (1985) used the electrophoretic technique to determine the differentiation in insect populations. Similar differences were found between susceptible and field larvae and also susceptible and field moths (Shekeban 2000).

In addition, Josephrajkumar and Subrahmanyam (2000)showed that the treatments with two insect growth regulators; namely, plumbagin and azadirachtin on cuticulogenesis at larval-pupal transformation reduced significantly the protein content. The cuticular protein profile of the treated insects showed variation in the number and prominence of bands compared to that of control insects.

Saleem and Rauf (2000) stated also that the combination of diflubenzuron + Cypermethrin showed increase in all the enzyme activities and some macromolecules. Soluble proteins and urea content were

decreased. Thus, it was suggested that synergism in combined applications with reference to all enzymes and various macromolecules would be possible.

Moreover, the effects of gamma hexachlorocyclohexane (gamma-HCH- lindane) and its combination with permethrin and Cypermethrin at the sublethal doses of LC_{10} and LC_{20} on some macromolecules in *T. castaneum* adult beetles were studied by Saleem *et al.* (2001) who concluded that the level of fructose, DNA and RNA remained unchanged for both doses. While the levels of glucose, lipids, cholesterol and soluble proteins remained undisturbed for LC_{10} dose, but LC_{20} dose their activites were decreased , Gamma – HCH + Cypermethrin at 10 + 1 and 20 +2 ppm depleted glucose, fructose, glycogen, soluble proteins and total proteins. However, DNA contents remained unchanged.

Ghoneim *et al.* (2001) determined the effects of exposure of *Oreochromis niloticus* to diazinon at the concentrations of 40,80 and 160 ppm for 1,2 and 4 weeks by estimating lipids profile, protein content, protein fingerprint, bilirubin, thyroid hormones and enzymes activity (glutathione transferase, creatine phosphokinase and acetyl cholinesterase) of treated fish serum . Dramatic changes were observed in most of the measured parameters regarding all the tested concentrations and the times of exposure. Serum protein fractionation by SDS-PAGE revealed the disappearance of certain protein fractions of the different concentrations of diazinon. Some of these fractions reappeared again after the recovery period, indicating the depressing effect of diazinon on the expression of certain genes.

In general, the present study showing that rate molecular markers are useful as indicators for the pest changes according to treatments differences.

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Table (1): The tested rates of Baythroid® alone and/or nutritive foliars in sequence during the growing cotton seasons of 2004 and 2005.

Treatments	Rate of application/l
Baythroid®/ Greenzit S.P$_{100}$/ Ascorbic acid	2.5 ml /0.2g /1 g
Baythroid® / Greenzit S.P$_{100}$	2.5ml/0.2g
Baythroid®/ Polymex / Ascorbic acid	2.5 ml/0.5 g/1g
Baythroid® / Polymex / Potasin-F	2.5 ml/0.5 g/5ml
Baythroid® /Greenzit N.P.K / Potasin-F	2.5ml/5 ml/1.5 ml
Baythroid® /Greenzit N.P.K / Neem oil	2.5ml/1.5ml/1ml
Baythroid®	2.5 ml

Cotton Seeds were treated with bio-fertilizer "Microbin®"

Table(2): Effect of the performed foliar sprays on the mean numbers of *Earias insulana* larvae per ten bolls of the bio-fertilized cotton plants throughout the growing cotton season of 2004.

Foliar Treatments and Rates of application/L	Mean number of larvae /10 bolls
Baythroid®/ Greenzit S.P$_{100}$/ Ascorbic acid 2.5ml/0.2g/1 g	1.27 ^{abcd}
Baythroid® / Greenzit S.P$_{100}$ 2.5ml/0.2g	0.87 ^{ab}
Baythroid®/ Polymex / Ascorbic acid 2.5 ml/0.5 g/1g	1.15 ^{abcd}
Baythroid® / Polymex / Potasin-F 2.5ml/0.5g/5ml	0.96 ^{abc}
Baythroid® /Greenzit N.P.K / Potasin-F 2.5ml/5ml/1.5ml	1.54^{d}
Baythroid® /Greenzit N.P.K / Neem oil 2.5 ml/1.5 ml/1.0 ml	0.81 ^{a}
Baythroid® 2.5ml	1.30 ^{bcd}
Untreated check	1.42^{cd}
F calculated = 4.99 F =	***

L.S.D = 0.328

Means followed by the same letter(s) are not significantly different at the 5% level

Table (3): Effect of the performed foliar sprays on the mean numbers of inspected *Earias insulana* larvae per ten bolls of the bio-fertilized cotton plants throughout the growing cotton season of 2005.

3Foliar Treatments and Rates of application/L	Mean number of larvae /10 bolls
Baythroid®/ Greenzit S.P_{100}/ Ascorbic acid 2.5ml/0.2g/1 g	0.63 [a]
Baythroid® / Greenzit S.P_{100} 2.5ml/0.2g	0.96 [ab]
Baythroid®/ Polymex / Ascorbic acid 2.5 ml/0.5 g/1g	0.90 [ab]
Baythroid® / Polymex / Potasin-F 2.5ml/0.5g/5ml	1.27 [b]
Baythroid® /Greenzit N.P.K / Potasin-F 2.5ml/5ml/.5ml	1.18 [b]
Baythroid® /Greenzit N.P.K / Neem oil 2.5 ml/1.5 ml/1.0 ml	1.0 [ab]
Baythroid® 2.5ml	0.87 [ab]
Untreated check	1.63[c]
F calculated = 7.6 F	***

L.S.$D_{0.05}$= 0.306

Means followed by the same letter(s) are not significantly different at the 5% level

Table(4): Effect of the tested foliar treatments on the cotton yield during the growing season of 2004.

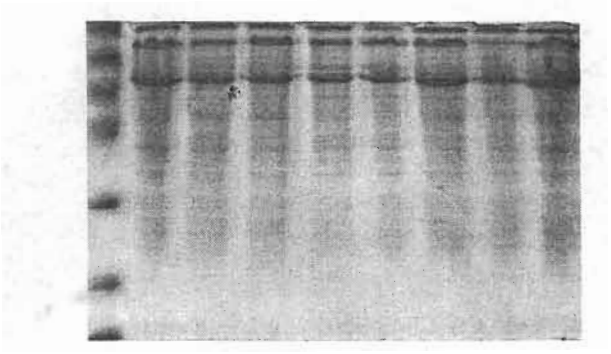
Treatments and Rates of application/L	2004 Cotton Yield/ Fed. Kg.	Kent.	* % Increase
Baythroid®/ Greenzit S.P$_{100}$/ Ascorbic acid 2.5ml/0.2g/1 g	914.4 Kg.	5.8(Kent.)	190%
Baythroid® / Greenzit S.P$_{100}$ 2.5ml/0.2g	982.6 Kg.	6.2(Kent.)	211.9%
Baythroid®/ Polymex / Ascorbic acid 2.5 ml/0.5 g/1g	840 Kg.	5.3(Kent.)	166.7%
Baythroid® / Polymex / Potasin-F 2.5ml/0.5g/5ml	810 Kg.	5.1(Kent.)	157.1%
Baythroid® /Greenzit N.P.K / Potasin-F 2.5 ml/5 ml/1.5 ml	765 Kg.	4.8(Kent.)	142.8%
Baythroid® /Greenzit N.P.K / Neem oil 2.5 ml/1.5 ml/1.0 ml	960 Kg.	6.09(Kent.)	204.7%
Baythroid® 2.5ml	750 Kg.	4.7(Kent.)	138.09%
Untreated check	315 Kg.	2(Kent.)	

*Expressed in % of increase than Untreated check, according to Hussein Nagwa M. *et al* (2002)

Table (5): Effect of tested foliar treatments on the cotton yield during the growing season of 2005.

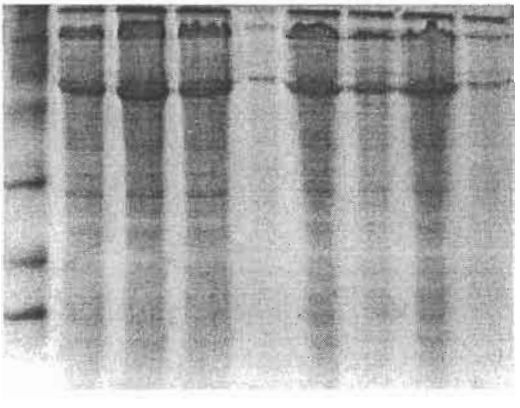
Treatments and Rates of application/L	2005		
	Cotton Yield/ Fed. Kg.	Kent.	* % Increase
Baythroid® / Greenzit *S.P$_{100}$*/ Ascorbic acid 2.5ml/0.2g/1 g	720 Kg.	4.5(Kent.)	71.4%
Baythroid®/ Greenzit S.P$_{100}$ 2.5ml/0.2g	696 Kg.	4.4(Kent.)	65.7%
Baythroid®/Polymex / Ascorbic acid 2.5 ml/0.5 g/1g	684 Kg.	4.3(Kent.)	62.8%
Baythroid® / Polymex / Potasin-F 2.5ml/0.5g/5ml	660 Kg.	4.1(Kent.)	57.1%
Baythroid®/Greenzit N.P.K /Potasin-F 2.5 ml/5 ml/1.5 ml	552 Kg.	3.5(Kent.)	31.4%
Baythroid®/ Greenzit N.P.K/Neem oil 2.5 ml/5 ml/1.5 ml	674 Kg.	4.2(Kent.)	60.4%
Baythroid® 2.5ml	420 Kg.	2.6(Kent.)	0
Untreated check	420 Kg.	2.6(Kent.)	

*Expressed in % of increase over untreated check, according to Hussein Nagwa M. *et al* (2002



1 2 3 4 5 6 7 8 9

Fig.(1):Polyacrylamide gel of total soluble protein patterns of untreated and treated larvae of *Earias Insulana*.
Lanes:1,Protein Marker;2, Baythroid®/Greenzit S.P$_{100}$/Ascorbic acid ;3 , Baythroid®/Greenzit S.P$_{100}$;4, Baythroid®/Polymex/Ascorbic acid;5, Baythroid®/Polymex/Potasin-F;6, Baythroid®/Potasin-F/Greenzit N.P.K ;7, Baythroid®/Greenzit N.P.K/Neem oil ;8, Baythroid® ;9 ,Untreated check(Season of 2004).



1 2 3 4 5 6 7 8 9

Fig.(2):Polyacrylamide gel of total soluble protein patterns of untreated and treated larvae of *Earias Insulana*.
Lanes:1,Protein Marker;2, Baythroid®/Greenzit S.P$_{100}$/Ascorbic acid ;3 ,Baythroid®/Greenzit S.P$_{100}$;4, Baythroid®/Polymex/ Ascorbic acid;5, Baythroid®/Polymex/Potasin-F;6, Baythroid®/Potasin-F/Greenzit N.P.K ;7, Baythroid®/Greenzit N.P.K/Neem oil ;8, Baythroid® ;9 ,Untreated check(Season of 2005).

Table (6): Densitometer analysis of total soluble protein SDS-PAGE showing band number& molecular weights in treated &untreated cotton spiny bollworm *Earias insulana*. in Season of 2004.

Band	M.W.	T_1	T_2	T_3	T_4	T_5	T_6	T_7	Control	Total
1	163.0	1	1	1	1	1	1	1	1	8
2	95.8	0	1	1	0	0	0	0	1	3
3	79.0	0	1	1	1	1	1	1	1	7
4	70.8	1	1	1	1	1	1	1	1	8
5	68.8	0	0	0	0	0	0	1	1	2
6	67.1	0	1	1	1	1	1	1	1	7
7	64.8	0	0	1	1	0	0	0	0	2
8	60.6	0	0	1	1	1	0	0	0	3
9	56.5	1	1	1	1	1	1	1	1	8
10	50.0	1	0	1	1	1	0	0	0	4
11	42.7	1	1	1	1	1	1	1	1	8
12	37.0	1	1	1	1	1	1	1	1	8
13	34.2	1	1	0	0	1	1	1	1	6
14	31.8	1	1	1	1	1	1	1	1	8
15	31.1	1	0	1	0	0	0	1	1	4
16	30.4	1	0	1	0	0	0	0	1	3
17	30.0	1	1	1	0	1	1	1	1	7
18	29.0	1	1	0	0	1	1	1	1	6
19	28.0	1	1	1	1	1	1	1	1	8
20	25.8	1	1	1	1	1	1	1	1	8
21	24.5	0	0	0	0	0	0	1	1	2
22	22.3	0	0	0	0	0	0	0	1	1
23	14.6	1	0	0	0	0	0	0	0	1
Total		15	14	17	13	15	13	16	19	122
Polymorphic bands		6	6	9	5	7	5	8	10	56
% Polymorphic bands		40	42.9	52.9	38.5	46.7	38.5	50	52.6	45.9
Specific bands		1	0	0	0	0	0	0	1	2

Table (7): Densitometer analysis of total soluble protein SDS-PAGE showing band number& molecular weights in treated &untreated cotton spiny bollworm *Earias insulana* (2005) .

Band	M.W.	T_1	T_2	T_3	T_4	T_5	T_6	T_7	Control	Total
1	204.3	1	1	1	1	1	1	1	0	7
2	140.8	0	1	1	1	0	0	1	1	5
3	103.9	1	0	0	0	1	1	1	1	5
4	77.6	1	1	0	1	1	1	1	1	7
5	70.3	1	1	1	0	1	1	1	1	7
6	69.2	1	1	1	1	1	1	1	1	8
7	64.3	1	1	1	1	1	1	1	1	8
8	52.1	0	1	1	0	1	1	0	1	5
9	44.2	1	1	1	1	1	1	1	1	8
10	38.6	1	1	1	0	0	1	0	1	5
11	35.5	1	1	0	1	1	1	1	1	7
12	33.2	0	1	1	1	0	1	1	0	5
13	32.3	1	1	0	0	1	0	0	1	4
14	31.0	0	0	1	0	0	1	1	1	4
15	30.4	1	1	1	1	1	1	1	0	7
16	29.7	1	1	1	1	1	1	1	1	8
17	28.7	0	0	0	1	0	1	1	1	4
18	28.0	1	1	1	0	1	1	0	0	5
19	25.8	1	0	1	0	1	1	1	0	5
20	24.5	0	1	0	0	1	1	1	0	4
21	21.0	1	0	0	0	1	1	1	0	4
22	19.4	1	1	1	0	1	1	1	1	7
23	16.6	0	0	1	0	1	1	1	1	5
24	14.3	1	1	1	0	1	1	1	0	6
25	11.2	1	1	1	0	1	1	1	0	6
26	9.0	0	0	0	0	1	0	0	0	1
Total		18	19	18	11	21	23	21	16	147
Polymorphic bands		14	15	14	7	17	19	17	12	115
% Polymorphic bands		77.8	79	77.8	63.6	81	82.6	81	75	78.2
Specific bands		0	0	0	0	1	0	0	0	1

الملخص العربى

الأثر السُمى والبيوكيميائى لبعض المُخصبات الورقية النباتية والمركبات البديلة على دودة اللوز الشوكية وعلاقته بالتفريد الكهربى لليرقات

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تهدف هذه الدراسة إلى تقييم تأثير المبيد الحشرى (بايثرويد) وبعض من مركبات المُخصبات الورقية التى تشمل العديد من العناصر الغذائية وبعض بدائل المركبات الكيميائية والتى أُستخدمت إما بصورة فردية أو تتابعية على النباتات المُسمدة حيوياً لصنف قطن "جيزة ٧٠ " خلال موسمى ٢٠٠٤-٢٠٠٥ وذلك ضمن برنامج المُكافحة المُتكاملة لدودة اللوز الشوكية بالإضافة إلى تحديد تأثيرها على المحتوى الكلى للبروتين فى اليرقات المُعاملة بأستخدام طريقة التفريد الكهربى.

وقد أظهرت النتائج الأتى :-

- أوضحت النتائج فى موسم ٢٠٠٤ أن إستخدام التتابعات الثنائية أو الثلاثية للبايثرويد مع جرينزيت أس – بى ١٠٠ أو مع جرينزيت ن – بو – فو / زيت النيم ظهور كفاءة فى خفض الإصابة بدودة اللوز الشوكية (٨١, ، ٨٧, يرقة /١٠ لوزات) ٠ بينما زاد مُعدل الإصابة بهذه الدودة فى باقى المُعاملات وتراوحت ما بين (٩٦, إلى ١,٣٠ يرقة /١٠ لوزات) ، فيما عدا أستخدام البايثرويد مع بوتاسين – أف و جرينزيت ن – بو – فو وكذلك فى النباتات الغير المُعاملة حيث إزدادت مُعدلات الإصابة إلى (٥٤, ١ ، ١,٤٢ يرقة /١٠ لوزات) .
- فى موسم ٢٠٠٥ أدى إستخدام البايثرويد بالتتابع مع جرينزيت أس- بى ١٠٠/ حمض الأسكوربيك وكذلك البايثرويد مُنفرداً إلى خفض كبير فى الإصابة بدودة اللوز الشوكية(٦٣, ، ٨٧, يرقة /١٠ لوزات) . مُقارنة بالنباتات الغير مُعاملة ١,٦٣ يرقة /١٠ لوزات) بينما إزداد مُعدل الإصابة بهذه الدودة فى باقى المُعاملات بمعدلات تراوحت ما بين ٩٠, إلى ١,٢٧ يرقة /١٠ لوزات .
- أدى إستخدام التتابعات الثنائية البايثرويد مع جرينزيت أس- بى ١٠٠ إلى زيادة الأنتاجية إلى ٦,٢ قنطار/فدان مُقارنة بالنباتات الغير مُعاملة (٤,٧ قنطار/فدان) وذلك فى موسم ٢٠٠٤؛ بينما فى

موسم ٢٠٠٥ أدى إستخدام البايثروبد بالتتابع مع جرينزيت أس- بى ١٠٠/ حمض الأسكوربيك إلى زيادة الأنتاجية ٤,٥ قنطار/فدان مقارنة بالنباتات الغير مُعاملة وكـذلك المُعاملـة بالبايثرويـد مُنفـرداً ٢,٦قنطار/فدان وذلك مرجعه إلى نقص أو زيادة أعداد يرقات الحشرة تحت الدراسة.

- دراسة توزيع الحزم البروتينية الكلية الذائبة الخاصة بأجسام اليرقات المعاملة بالمُعاملات المُختلفـة وكذلك اليرقات الغير معاملة عن طريق استخدام جهاز الفصل الكهربي بمادة البولي أكريلاميد جيل أوضح وجود اختلافات عديدة في توزيع هذه الحزم وكانت النتائج كما يلى:

١. أنخفض عدد حزم البروتينات من ١٩ حزمة في اليرقات الغير مُعاملة إلى(١٣ــ ١٧) حزمـة فى المُعاملات المُختلفة وذلك فى موسم ٢٠٠٤ .
٢. أنخفض عدد حزم البروتينات من ٢٣ حزمة في اليرقات المُعاملة بـ جرينزيت ن - بو - فو / زيت إلى(١١ــ ٢١) حزمة فى المُعاملات المُختلفة وذلك فى موسم ٢٠٠٥ .
٣. اختفاء حزم البروتينات المشاهدة أثبتت الفشل أو القصور في تخليق بعض البروتينـات حيـث يمكن أن يعزى ذلك إلى التأثيرات المطفرة (الطفرية) للمعاملات التي تـم اختبارهـا والتـي بالتبعية يمكن أعتبارها نتيجة الاختلاف أو الشذوذ في تركيب المادة الوراثية DNA .