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Alternative methods for the control of *Tuta absoluta*

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ABSTRACT: This article reviews the work done on the life history, damage symptoms, distribution, resistance and management of *Tuta absoluta* (*T.absoluta*) . It also gives an account of chemical control, biological control, and recent techniques of control. In addition, an experimental trials were carried out to evaluate the effectiveness of entamopathogenic fungi (*Beauveria bassiana, Trichoderma album)*, toxicity units of *Bacillus thuringiensis var. kurstaki* , *Alpha-Cypermethrin*, Electrochemical activated water *(EAW)* and Nano-particles of scilica *(SNPs)* as well as their combinations for controlling *T. absoluta*. In adults *T. absoluta*, 10 % mortality was occurred in the control group within of the test durations. *T.album* was more virulent (70%, mortality) than those of *B. bassiana* (60%, mortality), killing the treated larvae relatively quickly (LT_{50}: 3.5-5 days), in comparison with the *B. bassiana* (50 %, mortality) that began to affect the larvae only after 6 days (LT_{50}: 5.5.6.5 days). The larval mortality percentage of *T.absoluta* treated as newly hatched larvae with *EAW* + α-cyper + SNPs and *B. thuringiensis var. hsrstciki + EAW + α-cyper*. reached 90 %, followed by *EAW + α-cyper.* treatment 70 %) , while the value was 10% in the control. Percentages of hatchability of *T. absoluta treated* with *EAW + α-cyper* and *EAW+ α-cyper.+ SNPs* were 30 % for each. Meanwhile, the other treatments of *B. bassiana* , *Bacillus thuringiensis* and *T.album* were 80,72 and 50 %,respectively. On the other hand, percentage of hatchability of *T. absoluta* reduced to 25% in *EAW+ α-cyper + SNPs*.,while the value was 92% in the control. It is common knowledge that intensive chemical treatment leads to the development of resistance, and therefore alternative methods should be considered.

***Keywords*:** Alternative methods, *T.absoluta*, Control..

INTRODUCTION

Epidemiology. Tuta absoluta is one of the most important insect pests of tomato which posing a serious threat to tomato production across the Mediterranean. This pest is crossing borders rapidly and devastating tomato production substantially. *Tuta absoluta* is considered the major pest that attacks tomato in many countries. The newly introduced pest from South America is finding the shores of the Mediterranean a perfect new home where it can breed, between 10-12 generations in a year (Figure 1).

Figure 1. Epidemiology and distribution of The leaf miner Tuta absoluta finding the shores of the Mediterranean a perfect new home where it can breed, between 10-12 generations in a year

Figure 2. The leaf miner goes through six stages, namely egg, three larval stages, pupa and adult.a: the life cycle,b:Eggs on tomato leaf,c:Larval stages

Biology. The leaf miner goes through six stages, namely egg, three larval stages, pupa and adult (Figure 3). The adult leaf miners are small, yellow and black colored flies. The larvae form mines in the leaves of plants. Pupation takes place mostly in the soil. The larva feeds voraciously upon tomato plants, producing large galleries in leaves, burrowing in stalks, and consuming apical buds and green and ripe fruits. It is capable of causing a yield loss of 100% (1).

Figure 3. Cosmetic damage: leaves drying out or even early defoliation, secondary pathogens entering host plants through wounds made by the pest

Damage symptoms. Larvae cause mines. This can lead to cosmetic damage, leaves drying out or even early defoliation. The latter may affect the yield (Figure 3). Female adults cause feeding marks where they feed. This gives cosmetic damage to the plants. Indirect damage occurs when fungi or bacteria enter the feeding areas.

Infestation by *T. absoluta* has resulted in 50-100% losses in tomato. Yield and fruit quality are both significantly impacted by direct feeding of the leaf miner as well as secondary pathogens entering host plants through wounds made by the pest. Larvae penetrate the fruit, leaves, or stems of host plants, creating conspicuous mines and galleries and also allowing for invasion by secondary pathogens which may lead to fruit rot. Tomato plants may be attacked at any developmental stage. Infestation by the tomato leaf miner is easily detected on aerial buds, flowers, or new fruits. Chemical control is the main method of control for *T. absoluta*, but effective control is difficult to achieve because the larvae feed internally and develop resistance quickly.

Control. Some insecticides (indoxacarb, imidacloprid) and non-traditional methods (culture filtrate of *Bacillus thuringiensis, Artemisia cina* extract, clove oil and nanosilica) were evaluated against *T. absoluta* in tomato under greenhouse conditions (2). Nanosilica was the most effective treatment against *T. absoluta*

The sex pheromone for *Tuta absoluta* has been identified by researchers at Cornell University and has been found to be highly attractive to male moths. Pheromone lures are used extensively throughout Europe, South America, North Africa and the Middle East for the monitoring and mass-trapping of *Tuta absoluta* (3).

Biological controls can be used, including predatory bugs such as *Macrolophus pygmaeus* (commercially available as *Macrolophus caliginosus*) and *Nesidiocoris tenuis* which are large consumers of eggs. In the Mediterranean production areas, these two species naturally colonise tomato crops not sprayed with broad spectrum insecticides and are also released in greenhouse tomato crops.

Parasitoids are the most widely used natural enemies of *T. absoluta* in South America, where the pest originates, say researchers. In Europe, parasitoids have been found parasitising *T. absoluta* larvae in the Mediterranean area with at least two species of *Necremnus* identified in Spain and Italy. Regarding parasitoids of *T. absoluta* eggs, *Trichogramma acheae* has been identified as a potential biological control agent of the pest and is currently being released in commercial tomato greenhouses.
The effectiveness of entomopathogens against *T. absoluta* is poorly documented, with the exception of *Bacillus thuringiensis* var. kurstaki, which has been used extensively to control the pest in crops where IPM programmes based on biological control are applied.

Chemical control is difficult, say researchers, because the larvae live inside leaves, fruits and stems. Furthermore, pests such as *T. absoluta* , with a high reproductive capacity and very short generations, have an increased risk of developing resistance, say researchers. “It is therefore crucial to avoid systematic applications, and only apply treatments according to pest population density and crop damage following the recommendations of advisers.
Some populations of *T. absoluta* have developed resistance to organophosphate and pyrethroid pesticides (4). Newer compounds such as spinosadimidacloprid and *Bacillus thuringiensis* (5) have demonstrated some efficacy in controlling European outbreaks of this moth. Experiments have revealed some promising agents of biological pest control for this moth, including *Nabis pseudoferus,* a species of damsel bug (6).

Soley’s Tuta control mixture [extract of *Trichogramma pretiosum* (against adults) + fungi *Beauveria bassiana* (1 Billion cfu/gr ,gainst eggs and larvae) + fungi *Metarhizium anisopliae* (2 Billion cfu/gr,against eggs and larvae)] have demonstrated some efficacy in controlling European outbreaks.

MATERIALS AND METHODS

Experimental trials

In the present study, experimental trials were carried out to evaluate the efficacy of entamopathogenic organisms (*Beauveria bassiana, Trichoderma album*, toxicity units of *Bacillus thuringiensis var. kurstaki*) , *Alpha-Cypermethrin,* Electrochemical activated water *(EAW)* and Nano-particles of scilica *(SNPs)* as well as their combinations.

Agents

1. Protecto is a commercial formulation of *Bacillus thuringiensis var. kurstaki* and it is a product of Special Unit for Producing Bioinsecticides, Plant Protection Research Institute (PPRI), Agriculture Research Center (ARC). Egypt, with 32000 international toxicity units (spores and protein crystals) per mg. The active ingredient is 6.4% W-P- mid.
2. Biover a commercial formulation of *Beauveria bassiana* and it is a product of Special Unit of Producing Bioinsecticules. PPRI- ARC. Egypt, The international unit was 32, 000 viable spores per mg. The active ingredient was 10% W.P.
3. *Trichoderma album*. Source: Biozeid (Local) Company, Egypt. It contains 25 x 10^6 spores per mg.
4. *Alpha-Cypermethrin.* (Purity ≥ 98 %) was kindly supplied by Central Agricultural Pesticide Laboratory comprised of: (S)-alpha-cyano-3-phenoxybenzyl-(1R)-cis-3-(2,2dichlorovinyl) -2,2-dimethylcyclopropanecarboxylate and (R)-alpha-cyano-3-phenoxybenzyl-(1S)-cis-3-(2,2dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate Cypermethrin (0.001ml/L).
5. Electrochemical activated water *(EAW)*
6. *Hydrophobic nanosilica* (15–25 nm size range) at 100 ppm.

The Pest (Insect rearing)

The tomato leaf miner, *T. absoluta* samples from tomato growers. The pest was reared in the laboratory on tomato seedlings. The larvae were added with tomato seedlings in glass jars which were tightly closed with muslin.
If needed more tomato seedlings were added until pupation. When at least 5 pair of adults had emerged, they were put in glass cages (17 cm height and 7-12 cm in diameter) prepared with tomato seedlings as deposit of eggs and a piece of cotton saturated in a sugar solution10% to feed the moths and tightly closed with muslin.

Parameters were calculated using following formula

1. LT 50 = Lethal time of 50% of the population due to *Fungus or Bt* toxicity units were calculated with a probit analysis.
2. Mortality percentages: Were corrected according to Abbott s formula (1925 Mortality % = (No. of dead / Total No.) x 100. Corrected mortality % = (P - PO / 100 – P0) x 100 (P = Percent morality of treated; PO = Percent mortality of untreated).).
4. Hatchability percentage = (No.egg hatchability in check – No. egg hatchability in treatment / No. egg hatchability in check) x 100
5.

Agent test in bioassay and assay experiment

In this study two sets of experiments were conducted using the dipping method (7).

Procedures

First larval instars (L1) of Tuta .absoluta

A concentration of 10^8 spores .ml^{-1} of the strain of *fungus* or *Bt* toxicity units of *Bacillus thuringiensis +EAW+ α-cyper.* or *EAW+ α-cyper* or *EAW+ α-cyper.+SNPs* were assessed against the first larval instars (L1). The bioassay of experiment was performed with a group of 30 larvae at a temperature of 22-24 ºC. Bioassays were performed by dipping to first instar larvae in blastospore or combination suspensions for five seconds.

After allowing the excess liquid to drip off, the *T. absoluta* larvae were placed individually in rearing pots (cylindrical, inner diameter 45-100 mm, height 60 - 100 mm) filled 2/3 rd with sterile soil. Sliced potatoes serving as food were added to the peat. The larvae were checked at intervals for fungal infections. The peat was replaced as necessary. The lid of the rearing pots was perforated to facilitate aeration. One group of larvae was maintained as control just dipping in distilled water.

Eggs treatment

The eggs of *T .absoluta* on the tomato seedling were obtained from egg laying boxes were collected and put in jars following the treatment until hatching. The eggs were placed singly in plastic boxes or jar each containing 2.5 g sugar cane sawdust pretreated as follows: the sawdust was sprayed with fungal spore suspension (10^8 spores.ml^{-1}) of aqueous solution or *Bt* toxicity units of *Bacillus thuringiensis +EAW+ α-cyper.* or *EAW+ α-cyper.* or *EAW+ α-cyper.+SNPs* at a rate of 0.2 ml. gm^{-1} of sawdust and well mixed . The boxes with eggs were incubated for 10 days at 27 ºC in darkness. Egg hatching and mortality of emerging larvae were monitored during this period. The larvae that survived the treatment and of those in the control groups were examined. The egg bioassay was repeated three times with three different batches of eggs (10-24 eggs per treatment of each test group).

Adult treatment

Twenty pairs of adult *T. absoluta* were sprayed using a Badger 100 artists' airbrush to mist 0.4 ml of solution from a particular concentration of treatment and then placed inside the box for each treatment. Then they were put in glass cages (17 cm height and 7-12 cm in diameter) prepared with tomato seedlings.

After 1, 2, 4, 7 and 14 days of exposure, *T.absoluta* adults were observed in each treatment and combination to assess mortality.

RESULTS AND DISCUSSION

The results in Table 1 ,show that the larval mortality percentage of T.absoluta treated as newly hatched larvae with EAW+ α-cyper + SNPs reached 90 %, followed by B. thuringiensis var. hsrstciki +EAW+ α-cyper. 90 % and 70 % with EAW + α-cyper. treatment, while the value was 10% in the control (5) ; (8).

Percentage of hatchability of T. absoluta treated with EAW + α-cyper and EAW+ α-cyper.+ SNPs were 30 % for each. Meanwhile, the other treatments of B. bassiana, Bacillus thuringiensis and T.album were 80, 72 and 50 %,respectively. On the other hand,percentage of hachablity of T. absoluta reduced to 25% in EAW+ α-cyper + SNPs.,while the value was 92% in the control.

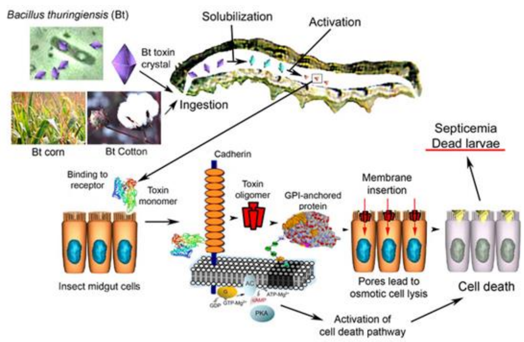

Figure 4. This multi-step toxicity process includes ingestion of the Cry protein by a susceptible insect, solubilization, and procesing from a protoxin to an activated toxin core in the insect digestive fluid. The toxin core travels across the peritrophic matrix and binds to specific receptors called cadherins on the brush border membrane of the gut cells. Toxin binding to cadherin proteins results in activation of an oncotic cell death pathway and/or formation of toxin oligomers that bind to GPI-anchored proteins and concentrate on regions of the cell membrane called lipid rafts. Accumulation of toxin oligomers results in toxin insertion in the membrane, pore formation, osmotic cell shock, and ultimately insect death. Whether oncosis, pore formation and/or both mechanisms are ultimately responsible for enterocyte death is still controversial. (9)

In adults, 10 % mortality was occurred in the control group within of the test durations. T.album tested strain was more virulent (70%, mortality) than those of B. bassiana (60%, mortality), killing the treated larvae relatively quickly (LT_{50}: 3.5-5 days), in comparison with the B. bassiana strain (50 %, mortality) that began to affect the larvae only after 6 days (LT_{50}: 5.5.6.5 days).

Percentage of hatchability of T. absoluta treated with EAW + α-cyper. and EAW+ α-cyper.+SNPs were 30 % for each. Meanwhile, the other treatments of B. bassiana , Bacillus thuringiensis and T.album were 80,72 and 50 %,respectively. On the other hand,percentage of hachablity of T. absoluta reduced to 25% in Env.+ α-cyper + SNPs.,while the value was 92% in the control.

Table 1. Efficacy of biopesticides, EAW + α-cyper.+SNPs; Bt toxin +EAW+ α-Cyper and EAW + α-Cyper.on T.absoluta

Biopesticides	Mortality of emerged larvae	Adult mortality (%)	Hatchability %
Bacillus thuringiensis	50	75	72
Beauveria bassiana	60	76	80
T.album	70	85	50
Bacillus thuringiensis +[a]Env.+ [b]α-cyper	85	90	30
[a]EAW.+ α-cyper	70	85	30
EAW.+ α-cyper.+[c] SNPs	90	25	30
Control	10	4	92

EAW :Electroactivated water[a]
Alpha-Cypermethrin[b]
Hydrophobic nanosilica (15–25 nm size range) at 100 ppm[c]

Figure 5. Photograph of control (a) and SNP treated dead T. absoluta larvae (b). The dead larvae and insects became extremely shrunk because of dehydration

In adults, 10 % mortality was occurred in the control group within of the test durations. T.album tested strain was more virulent (70%, mortality) than those of B. bassiana (60%, mortality), killing the treated larvae relatively quickly (LT_{50}: 3.5-5 days), in comparison with the B. bassiana strain (50 %, mortality) that began to affect the larvae only after 6 days (LT_{50}: 5.5.6.5 days).

On the other hand, the combinations : EAW + α-Cyper.+SNPs; Bacillus thuringiensis +EAW + α-Cyper ; EAW + α-Cyper. were efficient where they induced mortality percentages:90 (LT_{50}: 1-2 hrs),,85 (LT_{50}: 1-1. 5 hrs), and 70 (LT_{50}: 2-2.5 days),respectively.

One to 2 days later, the color of larvae killed by B. bassiana changed from sandy to dark pink. After development inside the larvae, both T.album and B. bassiana strains appeared to break through the cuticle and conidia emerged on the surfaces of the cadavers penetration by both fungi were identical and appeared 2 days PI.

Owing to the observed susceptibility of all the development stages of the T. absoluta to the entomopathogenic fungi under laboratory conditions, the practicability of achieving efficient control of T. absoluta in the field seems no problematic. The field efficacy of entomopathogenic fungi toward various pests depends on many factors, often related to the behavior of the insect host in its natural habitat. The soil is the natural habitat of fungi and, since the T. absoluta pupae inhabit the soil, it is theoretically possible to infect them with fungal spores by soil treatment.

IPM strategies are being developed in South America to control T. absoluta. Studies are being done on the use of synthetic sex pheromones in order to monitor population levels and trigger applications of chemicals (3). Various active substances are effective and can be used in combination with biological control agents. Concerning chemical control, several treatments are required per growing season and it must be noted that a decrease of the efficacy of products used against T. absoluta has been observed since the 1980s in tomato crops. Resistance to some insecticides has been reported in several countries, for example to abamectin, cartap and permethrin in Brazil (Siqueira infested plants and of post-harvest plant debris, etc.) (3). Finally, the susceptibility of tomato cultivars to T. absoluta varies and plant resistance is being investigated.

Concerning adults, 12 % mortality was occurred in the control group within of the test durations. T.album tested strain was more virulent than those of B. bassiana, killing the treated larvae relatively quickly (LT_{50}: 4-5 days), in comparison with the B. bassiana strain that began to affect the larvae only after 6 days (LT_{50}: 6.7 days).

One to 2 days later, the color of larvae killed by B. bassiana changed from sandy to dark pink. After development inside the larvae, both T.album and B. bassiana strains appeared to break through the cuticle and conidia emerged on the surfaces of the cadavers penetration by both fungi were identical and appeared 2days PI (8).

Adults killed by the fungus did not change color, whereas dead adults in the control treatment darkened. After incubation of cadavers under moist conditions, fungi emerged on the dorsal and ventral surfaces of the weevil and formed conidiophores with conidia.

CONCULSION

It is common knowledge that intensive chemical treatment leads to the development of resistance, and therefore alternative methods should be considered. To control the pest effectively it is critical to combine all the control measures available and not to rely only on insecticide sprays. Various active substances are effective and can be used in combination with biological control agents. It is very important to pay attention to the side effects of pesticides on natural enemies, especially predatory bugs. As these individuals often have a slow establishment process, the insecticide should be selected carefully, especially in the early growth stages of the crop.

REFERENCES

Derbalah AS, Morsey SZ and El-Samahy M. 2012. Some recent approaches to control Tuta absoluta in tomato under greenhouse conditions.African Entomology (20) 1:27-34.

Gonzalez-Cabrera J, Molla O, Monton H and Urbaneja A. 2011. Efficacy of *Bacillus thuringiensis* (Berliner) for controlling the tomato borer, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). Biocontrol 56, 71–80.

Goettel MS and Inglis DG. 1997. Fungi: Hyphomycetes. In: Manual of techniques in insect pathology. Lacey LA, edited by Academic Press, London, UK, pp, 213-249.

http://web.utk.edu/~jurat/Btresearchtable.html

Kaoud HA, Saeid S, EL-dahshan AZ and El-Behary AM. 2013. New Methods for the Control of Lesser Grain Borer, Rhyzopertha Dominica. International Journal of Engineering and Innovative Technology (IJEIT) Volume 3(4):285-289.

Lietti MMM, Botto E and Alzogaray RA. 2005. Insecticide Resistance in Argentine Populations of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). Neotropical Entomology 34(1):113-119.

Molla O, Gonzalez-Cabrera J and Urbaneja A. ,2011. The combined use of *Bacillus thuringiensis* and *Nesidiocoris* tenuis *against the tomato borer* Tuta absoluta. Biocontrol 56, 71–80

pablaza J. 1992. La polilla del tomate y su manejo. Tattersal 79, 12–13.

Salas J. 2004. [Capture of *Tuta absoluta* in traps baited with its sex pheromone.] *Revista Colombiana de Entomología* 20, 75–78 (in Spanish).