Local bacterial isolates as entomopathogenic agents against the citrus flower moth, *Prays citri* Miller (Lepidoptera, Hyponomeutidae) in lime orchards at north Delta region, Egypt

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Abstract

Three species of entomopathogenic bacteria were isolated identified and tested from citrus flower moth larvae (CFM). The effect of Entomopathogenes on the citrus flower moth under laboratory conditions was studied. Also, evaluation the efficacy of local bacterial isolates on *P. citri* population under field conditions was studied. Statistical analysis indicated that *B. sphaericus* had the highest pathogenicity to CFM, while *B. subtilis* ranked second in the order of activity, *B. thurengeinsis* had the lowest entomopathogenic bacterium.
Key words: *Bacillus thurengeinsis, B. subtilis* and *B. sphaericus, Prays citri*

Introduction

Citrus crop consider one of the most important fruit crops in Egypt. Many pests are attacking citrus tress, some of these cause serious damage, hence affecting quantity and quality of the fruits and cause economic loss in the crop. The citrus flower moth (CFM), *Prays citri* Millière (Lepidoptera: Yponomeutidae), is regarded as a key pest of citrus in parts of the Mediterranean, including Portugal and Spain. In addition, it has been reported to attack other Rutaceae and species of Sapotaceae (*Manilkara zapota*, *Casimiroa edulis*) and Oleaceae (*Ligustrum lucidum*). Damage may vary depending on the citrus crop attacked. Its major host is *Citrus aurantiifolia* (lime), but damage can occur also on *Citrus limon* (lemon) and occasionally to other citrus fruits, including sweet orange (*Citrus sinensis*). The larva feeds both internally and externally on flowers, flower buds and fruits; it can also feed on leaves and young shoots (**Carimi *et al.*, 2000**, **EFSA, 2008**). In Egypt, this insect has 9-11 generations with two main peaks; the first is during April, May and June while the heaviest on lime trees occurred in Septemper, October and November **(Shehata, 1982 and El-Dessouki, *et al* 1987).**

Previous information on *P. cirri* control recorded in some available literature indicated that chemical control was the most measures using against this pest in Egypt (**Shehata, 1982 and Atrm *et al.*, 1992**). According to **Smith (1986)**, the application of insecticides is frequently followed by recurrent infestations in addition to several problems such undesirable chemical residuals, environmental pollution, insecticides resistance. So, although the application of insecticides which used to control this insect pest in Egypt, it still causes considerable damage to its host plants. Therefore, during the past decade, intensive efforts have been done in Egypt to improve the biological control and to devise an integrated control program for citrus pests. The search for new microbial agents for pest control is one of the most pressing needs in the field of biological control. Therefore, isolation of more local entomopathogens that would be more adapted to the local pest hosts and possesses greater insecticidal activities or broader host range (**Abd-Elazim *et al.*, 1991; Osman, 1992** and **Keller, 1998**).

Endospores forming bacteria are resilient structures capable of surviving desiccation, heat, oxidizing agents, and UV and γ radiation. These qualities confer exceptional ecological advantages to bacilli and allow for long-term storage and

relatively easy commercialization of *Bacillus*-based products. **(Collins and Jacobsen, 2003)**. *B. thuringiensis* isolated from the larvae of *Prays oleae* for the first time in Egypt **(Nasr et al, 2002).** *B. thuringiensis* has been the leading biopesticide against lepidopterous pests since 1959. Since the 1990s, the use of bacteria endowed with entomocidal properties for biological control of insect pests has risen. Endotoxins from the spore-forming bacterium, *Bacillus thuringiensis*, are the most valuable biopesticides used currently in commercial agriculture, forest management and mosquito control **(Tounsi *et al*., 2007).** Today a variety of entomopathogens are used for the control of invertebrate pests in glasshouse and row crops, orchards and ornamentals. B. *thuringiensis* is considered an effective biological control agent. This bacterium produces a very specific insecticidal proteins with toxicity to several orders of insect species **(Navon, 2000; Huang *et al*., 2007 and Rodriguez *et al*., 2009**).

B. thuringiensis produce this proteins during sporulation which exhibit a wide variety of Mediterranean flour moth such as lepidopteran, coleopteran and dipteran insects species. These proteins was known as crystal protein (cry protein); crystal toxin (cry toxin); Delta-endotoxins (δ-endotoxins) and protoxins. There were another kinds of proteins namely the vegetative insecticidal proteins (Vip protein), produced from *B. thuringiensis* during the vegetative stage of its growth **(Lacey *et al*., 2001; Tounsi *et al*., 2006a and Tounsi *et al*., 2006b).** These toxins account for up to 30% of the total protein content of *B. thuringiensis* (**Lacey *et al*., 2001).** More than 300 cry genes have been sequenced and the cry proteins are classified into at least 49 groups organized into subgroups according to the percentage identity of their amino acid sequences **(Martins *et al*., 2007**).

So, the objective of this study was aimed to collect, isolate and identify the local entomopathogeneic bacteria associated with the citrus flower moth, *P. citri* under laboratory conditions. In addition, the efficiency of these entomopathoges against *P. citri* under field conditions.

Materials and methods

I- Field experiments :

1. Samples collection :

The present experiments were conducted in Damietta Governorate, Kafr-Saad district, throughout two successive months (from the 1st of March till the 30th of April 2010). One feddan had been selected to collect and isolate the local entomopathgenic bacteria which associated with the citrus flower moth (CFM), *Prays citri* Millière in lime orchards.

Samples were collected weekly during two successive months. Each sample consisted of 250 flowers from the experimental area. Samples were covered with polyethylene bag in the field and then pulled up and transferred to the laboratory for examination.

To collect and isolate pathogeneic bacteria, living and dead individuals of *P. citri* which show general infected symptoms were distinguished and put into sterilized tubes. The collected tubes were transferred to the Microbiological lab, (Microbiology Department, Faculty of Agriculture, Mansoura University) to isolate and identify the bacterial species presented on the pest.

2- Isolation and identification of microorganisms :

The insect was crushed and 10 ml of sterile tap water was added. The suspension was shacked well for 10 min and serially diluted in sterile tap water and the dilutions from 10^{-1} to 10^{-6} were plated on nutrient agar media. After incubation at 30±1°C for 2 days, plates were examined and the developed colonies were transferred on nutrient agar slants for subsequently testes. Nutrient agar medium was (g/l) :

peptone 5.0; beef extract 3.0 agar agar 20.0 and pH was 7.0. This medium was used for maintenance of all isolates. The isolates were purified and identified in Microbiology Department, Damittae Faculty of Agriculture, Mansoura University according to **(Holt *et al*., 1994)**

Bacteria were scraped off from the agar surface and saline water suspensions (NaCl 0.85%) were observed under light microscope (1,000x) to confirm the presence of parasporal crystals, a typical characteristic of *B. thuringiensis* **(Cavados *et al*., 2001 and Ozturk *et al*., 2008).**

3. Insect culture :

To prepare a laboratory culture from *P. citri*, larvae of *P. citri* were collected from infested lime flowers in lime orchards at Damietta Governorate. Larvae placed in Petri-dishes with fresh lime flower until pupation. The method of **El-Dessouki *et al.,*(1987)** was used for rearing. Five pairs (as replicates) of newly emerged adults moths (males + females) were released in separate cages provided with 10% sucrose solution for feeding moths and a fine lime branch contained many flowers. Newly deposited eggs were transferred to Petri-dishes with fresh flowers.

4. Pathogenicity of bacterial isolates:

Preparation of inoculums : The bacterial growths on the nutrient agar slants were scraped, using 5 ml sterile tap water, then transferred to a flask containing 50 ml sterile nutrient broth (**Elcin, 1995**). The resulting cell suspensions was calculated using plate count agar method according to **Post (1988).** The inocula were prepared using this suspension and were adjusted to 10^4 , 10^5 and 10^6 CFU/ml (colony forming unit) by dilution of sterile tap water (**Collins *et al*., 2003**).

To evaluate the pathogenicity of bacterial isolates on *P. citri*, two laboratory experiments were done:

The first experiment (primary experiment) : Ten larvae of *P. citri* (mixture of different instars) were put in a Petri-dish (9 cm in diameter) containing small branches of lime with enough numbers of flowers was sprays with one bacterial suspension (approximately 2 cm) The concentration of all inocula were $1x10^6$ CFU/ml. Each treatment was replicated five times as replicate. Additional five Petri-dishes were sprayed with nutrient broth only as control. Larvae were held until pupation, adult emergence, percentage of adult emergence and malformation moths were calculated.

The second experiment : Each inoculum of tested bacteria was used with three concentrations against each stage of *P. citri* larvae (1^{st}, 2^{nd}, 3^{rd} and 4^{th} instar). Three different concentrations of tested bacteria were used as an inoculum and it were $1x10^6$, $1x10^5$ and $1x10^4$ CFU/ml. Ten larvae of each stage were put in a Petri-dish (9cm. in diameter) containing small branches of lime with enough numbers of flowers was sprays with one concentration (2cm/ Petri-dish approximately) of a bacterial suspension. Each treatment was replicated five times as replicate. Additional five Petri-dishes were sprayed with a diluted nutrient broth medium only as control under laboratory conditions (Max.temp 30, Mintemp. 17, Max R.H. 85, Min. R.H 45). Larvae were held until pupation, adult emergence, percentage of adult emergence and malformation moths were calculated..

5. Evaluation the efficacy of local bacterial isolates on *P. citri* population under field conditions :

To evaluate the efficacy of the local bacterial isolates on *P. citri* population, an area of about 1000 m^2 of a lime orchard located at Damietta Governorate, Kafr-Saad district was selected and divided into four plots (three plots for the isolated bacteria and the other one for control treatment). During the main flowering period of lime trees (from the 3^{rd} till the 15^{th} of June), infested trees were selected and marked. The

highest concentration of each isolated bacteria agent ($1x10^6$ CFU/ml) was applied against ten trees (as replicates) in one plot. For each treatment, five branches were taken from each replicate (tree) after 3, 6, 9 and 12 days of treatment and then covered with polyethylene bag in the field and then pulled up and transferred to the laboratory for examination. The number of *P. citri* larvae were counted and recorded; in addition, infestation percentages were calculated. Reduction percentages in both population and infestation percentages were estimated according to **Henderson-Tilton's formula (1955)**. Statistical analysis was done by using one way ANOVA (**CoStat, 1990**); in addition to the simple regression analysis also was done.

RESULTS AND DISCUSSION

1-Characterization and identification of bacterial isolates:

Identification testes of the isolates showed that all isolates were bacilli, positive to Gram stain, endospore forming, motile, catalase positive, Methyl red test (M.R.) positive, oxidase positive, hydrolyzing gelatin, indole negative, Voges Proskauer test (V.P.) positive, and citrate utilization positive.

One isolate (A1) hydrolyzing starch and casein, produced acid from glucose, arabinose, xylose, manitol, not produced gas from glucose, From these results, these isolates were identified and designated as *B. subtilis* **(Holt *et al*., 1994).**

The second isolate (A2) hydrolyzing starch and casein, produced acid from glucose, not from manitol, xylose, arabinose, not produced gas from glucose. The presence of parasporal crystals was confirmed. These isolates were identified and designated as *B. thuringiensis* **(Holt *et al*., 1994)**.

The third isolate (B) hydrolyzing starch negative, nitrate reduction negative. These isolates were identified and designated as *B. sphaericus* **(Holt *et al*., 1994)**.

2- Entomopathogenes on the citrus flower moth under laboratory conditions :

2-1 primary experiment

Data illustrated in Figure (1) showed the pathogenicity of microbial isolates to the larval stage of CFM feed on treated flower with the isolated bacteria under laboratory conditions. The pathogenicity of each pathogen suspension was evaluated under laboratory conditions against the mixture of larval instar of CFM. The obtained results as shown in Figure (1) indicated that there are significant differences were apparent for bacterial infection between the inoculated and control larvae.

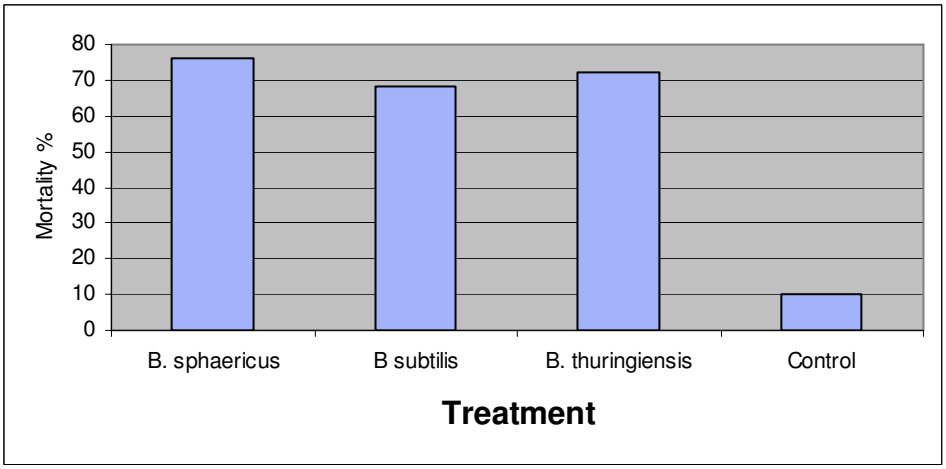

Figure (1). Pathogenicity of the isolated bacteria to *P. citri* larvae under laboratory conditions (L.S.D.$_{P=5\%}$ = 14.5 & $_{P=1\%}$= 20.0).

Table (1). Effect of the entomopathogenic bacteria on the adult stages of *P. citri* treated as 1st , 2nd , 3rd and 4th instar larval under laboratory conditions..

Treatment	Concentration (CFU/ml)	Instar				L.S.D. (P=5%)	L.S.D. (P=1%)
		1st	2nd	3rd	4th		
		Mortality ±SD					
B. sphaericus	10^6	100±0.0	90±4.5	94±8.9	88±13.0	15.7	21.7
	10^5	94±8.9	90±12.2	72±13.0	82±8.4	14.5	20.0
	10^4	94±8.9	78±11.0	56±11.4	60±21.2	18.2	25.8
B. subtilis	10^6	100±0.0	96±5.5	98±4.5	58±14.8	11.0	15.2
	10^5	96±5.5	90±14.1	74±11.4	36±5.5	13.2	18.2
	10^4	84±15.2	76±11.4	64±15.2	36±15.2	19.2	26.5
B. thurengeinsis	10^6	80±21.2	80±7.1	90±12.2	72±8.4	18.0	24.8
	10^5	76±11.4	70±26.5	90±10.0	66±20.7	24.7	34.7
	10^4	66±11.4	56±20.1	80±12.3	60±10.0	21.8	30.1
Control		12±8.4	6±8.9	6±8.9	6±5.5	10.8	14.9
L.S.D. (P=5%)		14.0	20.0	14.2	17.1		
L.S.D. (P=1%)		18.74	26.72	19.05	22.95		

Data presented in Table 1 show that, the toxic effect of different in column densities of the bacterial species on CFM larvae (four larvae instars) revealed that, all of bacterial treatments significantly recorded high percentages of mortality in comparison with the control treatment (distilled water).

One of the most important economic aspects of pest management with *B. thuringiensis* is the use of the microbe against young larvae, preferably neonates, because it has been shown in laboratory and field bioassays that third instar larvae of Lepidopteran and Coleopteran are less susceptible to the *B. thuringiensis* products than younger larvae. Thus, the costs of controlling 1st instar larvae can be a fraction of that of controlling older larvae. These differences in larval susceptibility to *B. thuringiensis* are probably aspects of a more general phenomenon among lepidopteran and coleopteran pests. Furthermore, the initial nibbling of *B. thuringiensis* by neonate larvae is followed by cessation of feeding and gut paralysis within minutes, so that there is negligible damage to plants, whereas mature larvae feeding on *B. thuringiensis*-treated plants may still cause sufficient damage to the crop to reduce the quality of the agricultural product. Also, mature larvae may recover from the *B. thuringiensis* intoxication, and even complete the developmental cycle; this situation can be aggravated by incomplete spray coverage, by rapid degradation of the crystalline toxin by UV radiation, or by washing off of the *B. thuringiensis* spray by rain or overhead irrigation **(Navon, 2000).**

Figure (2) shows the Log. of the concentration and mortality % as 1st, 2nd, 3rd and 4th instar larvae under laboratory conditions.

Data in Table 2 show that, the entomopathogenic bacterial species exhibited some morphological changes in CFM adult stages, when CFM treated as larval stage, bacterial suspension caused disturbance in the formed

emerged adults (where they had abnormal shape and crinkle wings in form). The malformation effect of different inoculum densities of the bacterial species on CFM adults stages under laboratory conditions was summarized in Table 2. In the 1st and 2nd instar larval and treatment *B. thurengeinsis* recorded relatively higher malformation percentages on CFM adults than *B. subtilis* and *B. sphaericus* . In the 3rd and 4th instar larval *B. subtilis* recorded relatively higher malformation percentages on CFM adults than *B. thurengeinsis* and *B. sphaericus.* The percentage of malformation caused by each entomopathogen increased as its concentration increase.

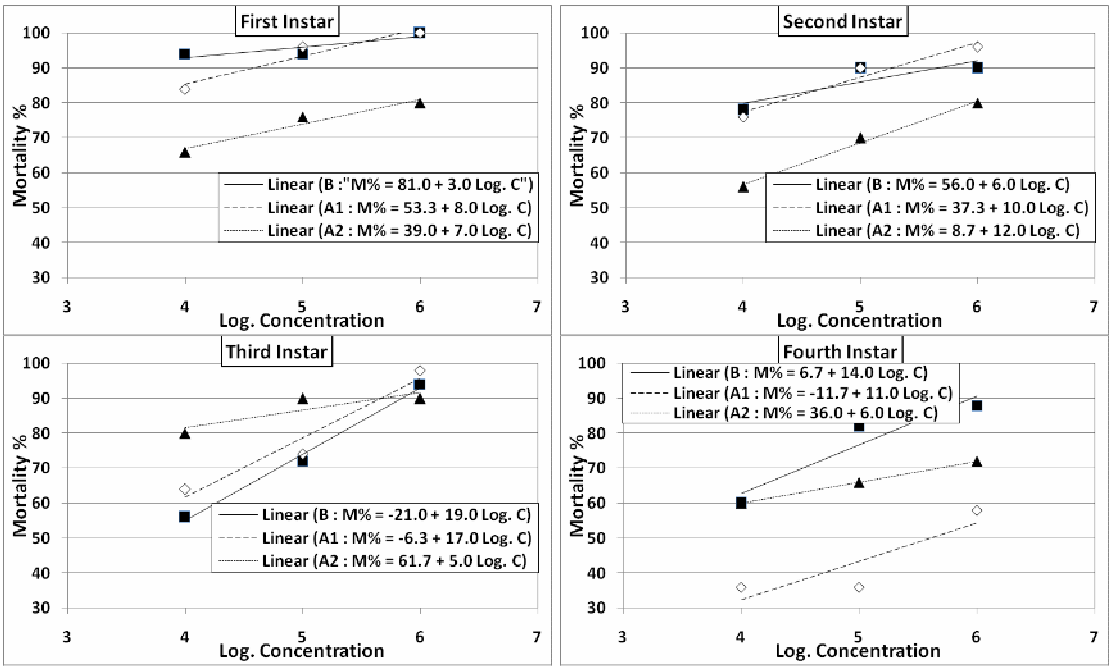

Figure (2). The relationships between the concentrations of the local isolated entomopathogenic bacteria and mortality percentages of *P. citri* treated as 1st, 2nd, 3rd and 4th instar larvae under laboratory conditions.

5. Evaluation the efficacy of local bacterial isolates on *P. citri* population under field conditions

As shown in Table 3, the mean reduction percentages of *p. citri* in treated plots with *B. subtilis*, A2 and B1 were 73.15±7.28, 69.13±4.06 and 79.2±1.7 %, respectively. While the mean reduction percentages of infestation by *p. citri* larvae in treated plots with *B. subtilis*,A2 and B1 were 70.82±4.01, 67.37±6.23 and 77.66±2.33 % during respectively. Statistical analysis indicated that *B. sphaericus* had the highest pathogenicity to CFM, while *B. subtilis* ranked second in the order of activity, *B. thurengeinsis* had the lowest entomopathogenic bacterium.

Table (2). Malformation percentage of *P. citri* moths treated as 1st, 2nd, 3rd and 4th instar larvae under laboratory conditions.

Treatment	Concentration (CFU/ml)	Instar				L.S.D. (P=5%)	L.S.D. (P=1%)
		1st	2nd	3rd	4th		
B. sphaericus	10^6	4±5.5	6±8.9	6±8.9	10±10.0	11.4	15.7
	10^5	2±4.5	8±4.5	4±5.5	18±8.4	7.9	10.9
	10^4	2±4.5	2±4.5	6±5.5	8±8.4	7.9	10.9
B. subtilis	10^6	6±8.9	10±7.1	10±10.0	16±11.4	12.7	17.5
	10^5	2±4.5	8±11.0	12±8.4	12±8.4	11.2	15.4
	10^4	10±10.0	6±5.5	12±13.0	12±8.4	12.9	17.8
B. thurengeinsis	10^6	10±14.1	10±7.1	6±5.5	16±5.5	11.8	16.3
	10^5	10±7.1	10±10.0	8±8.4	6±5.5	10.6	14.6
	10^4	10±10.0	6±5.0	4±5.5	10±7.1	9.7	13.4
Control		2±4.5	0.0±0.0	0.0±0.0	0±0.0	3.0	4.1
L.S.D. (P=5%)		10.2	9.0	10.0	10.1		
L.S.D. (P=1%)		13.7	12.1	13.35	13.5		

Table (3). Reduction percentages in *P. citri* population and infestation after 3, 6, 9, 12 days of treatment with local isolates of B, A1 and A2 under field conditions.

Duration after treatment	Reduction percentages in					
	P. citri population			Infestation with *P. citri*		
	B. subtilis	*B. thurengeinsis*	*B. sphaericus*	*B. subtilis*	*B. thurengeinsis*	*B. sphaericus*
3 days	62.5	65.6	78.1	68.7	64.4	77.0
6 days	77.8	68.9	80.0	74.5	67.3	79.4
9 days	78.1	75.0	81.3	74.7	77.0	79.9
12 days	72.5	67.5	77.5	66.9	63.3	74.9
Mean ± SD	73.2±7.3	69.1±4.1	79.2±1.7	70.8±4.0	67.4±6.2	77.7±2.3
L.S.D. (P=5%)	7.7			7.2		

Discussion

Bacterial toxins and mode of action:

B. thuringiensis Crystal (Cry) and Cytolitic (Cyt) protein families are a diverse group of proteins with activity against insects of different orders Lepidoptera, Coleoptera, Diptera and also against other invertebrates such as nematodes. These toxins are highly specific to their target insect, are innocuous to humans, vertebrates and plants, and are completely biodegradable. Therefore, *B. thuringiensis* is a viable alternative for the control of insect pests in agriculture and of important human disease vectors**.** *B. thuringiensis* Cry and Cyt toxins belong to a class of bacterial toxins known as pore-forming toxins (PFT) that are secreted as water-soluble proteins undergoing conformational changes in order to insert into, or to translocate across, cell membranes of their host. There are two main groups of PFT: (i) the ά-helical toxins, in which ά-helix regions form the trans-membrane pore, and (ii) the β-barrel toxins, that

insert into the membrane by forming a β-barrel composed of β-sheet hairpins from each monomer. The first class of PFT includes toxins such as the colicins, exotoxin A, diphtheria toxin and also the Cry three-domain toxins. On the other hand, aerolysin, ά-hemolysin, anthrax protective antigen, cholesterol-dependent toxins as the perfringolysin O and the Cyt toxins belong to the β-barrel toxins. In general, PFT-producing bacteria secrete their toxins and these toxins interact with specific receptors located on the host cell surface. In most cases, PFT are activated by host proteases after receptor binding inducing the formation of an oligomeric structure that is insertion competent. Finally, membrane insertion is triggered, in most cases, by a decrease in pH that induces a molten globule state of the protein **(Bravo *et al*., 2007).**

Delta-endotoxins encoded by cry genes have been classified as cry protein, depending on their amino acid sequences and host specificity. *B. thuringiensis* produces crystal proteins toxic to Lepidopteran insect larvae. The crystals of *B. thuringiensis* are composed of three kinds of protein **(Tounsi *et al*., 2005).** Almost, all cry proteins are activated to 60–70 kDa protease resistant proteins. The proteolytic activation of cry toxins involves the removal of a short N-terminal peptide of 25–30 amino acids for Cry1 toxins, 58 residues for Cry3A and 49 amino acids for Cry2Aa. In the case of long Cry protoxins, approximately half of the remaining proteins are removed from the C-terminus. The mixture of toxins have shown cell damage and toxin binding to the midgut epithelial cells. The in vivo activation process is not clear and could vary because of differences in insect midgut proteases **(Monnerat *et al*., 2007; Rodriguez *et al*., 2009 and Dammaka *et al*., 2010).**

After the ingestion and solubilization of cry proteins (protoxins) in the alkaline midgut, cleavage by gut proteases produces a smaller 60- to 65-kDa activated protein that recognizes specific binding sites at the brush border membrane surface of the epithelial columnar cells lining the gut lumen. The next steps are pore formation, membrane transport disruption, and cell lysis leading ultimately to insect death. Independently of their size, activation by proteolytic digestion is a crucial step in the mode of action of the cry proteins. the mode of action of Vip toxin attacks the midgut larvae causing disruption of epithelial cells and leakage of material in the lumen **(Dammaka *et al*., 2010 and Lacey *et al*., 2001).**

B. subtilis isolates have shown the capacity to biological control of insect pests. *B. subtilis* also produce a range of antibiotic compounds that are inhibitory to fungi, bacteria and insects **(Osborn *et al*., 2002** and **Collins and Jacobsen, 2003**).

B. sphaericus is now commercially produced and has some advantages over *B. thuringiensis* in that it is more persistent in polluted habitats and may recycle under certain conditions, but has a narrower host range **(Elcin, 1995 and Lacey *et al*., 2001**). *Bacillus thuringiensis* (*Bt*) and *Bacillus sphaericus* (*Bs*) are two safe biological control agents. They have attracted considerable interest as possible replacements for the chemical insecticides **(El-Bendary, 2006).**

Some strains of *B. sphaericus* are toxic towards larvae and can be used as biological control agents. These entomocidal *B. sphaericus* strains can synthesize two different types of toxin: the binary toxin (Bin) and a mosquitocidal toxin (Mtx). All strains which synthesize the binary toxin are considered as high-toxicity strains, while low-toxicity strains synthesize only Mtx or neither toxin **(Silva *et al*., 1999).**

The examination of *B. subtilis* by the electron microscope contained large amorphous inclusions, whereas *B. sphaericus* contained crystals indistinguishable from those of *B. sphaericus*. These results suggest the presence of a factor absent in *B. subtilis* necessary for ordered aggregation of the 51- and 42-kDa proteins. Such a factor must also be present in *B. thuringiensis*, since crystalline inclusions of the *B. sphaericus* toxin are formed in this species. It should be noted that in the case of lepidoptera-active toxins of *B. thuringiensis*, crystalline inclusions are produced by *B. subtilis* (**Baumann *et al*., 1991)**

The pathogenic effect of Bactospeine, *B. thurngiensis* (Berliner) was higher than that of Bio-fly, *Beauveria bassiana* in spite of the latter was applied with higher doses on *P. citri***.** Two sprays of Bactospeine are adequate effective to reduce the population of *P. citri* in lime orchards during the main flowering period of trees without need of chemical insecticides **(Shetata and Feebv ,1998).** Agerin (*B. thuringiensis*) or the egg parasitoid *T. evanescenare* suitable candidates for the control of *P. citri* in lime orchards within an integrated pest management system in Egyp, the percent reductions in the larval infestation of *P*. citri (compared to the untreated trees) were 62.2, 76.4 and 78.3 in 2002 and 65.9, 80.4 and 75.9 2003 after application of *T. evanescenare,* Agrin and Ethion, respectively (**Abo-Sheaesha and Agamy, 2004**). 77.8, 91.2 and 100 % mortalities in *P. citri* larvae on artificially infested treated with suspensions of *B. thurngiensis* containing $2.5x10^8$, $2.5x\ 10^9$ and $3.3\ x10^9$ spores/ml. in the field, respectively (**Giammanco *et al*, 1966**).

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عزلات بكتيرية محلية كممرضات لفراشة أزهار الموالح (*Prays citri*) (رتبة غمدية الأجنحة)
على أشجار الليمون بشمال الدلتا – مصر

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الملخص العربى

تم عزل وتعريف ثلاثة أنواع من المسببات المرضية البكتيرية على أنها *Bacillus thurengeinsis* و*B. subtilis* و *B. sphaericus* وذلك من يرقات فراشة أزهار الموالح من منطقة كفر سعد بمحافظة دمياط وتم إختبار قدرة هذه البكتريا المرضية على الحشرة فى تجارب معملية وحقلية وكان من أهم النتائج حدوث موت وتشوهات مورفولوجية فى الطور الكامل لفراشة أزهار الموالح وكانت أفضل المعاملات هى المعامل بـ *B. sphaericus* .