



Efficiency of Three Bacterial Strains Against *Tetranychus urticae* Koch (Acari: Tetranychidae) Under Laboratory Conditions

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Abstract

This study was conducted to evaluate the efficiency of three bacterial strains *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Lysinibacillus spaericus* against adult females of *Tetranychus urticae* Koch using spraying and dipping techniques with concentrations (10^1 , 10^3 , 10^5 , 10^7 cfu. ml⁻¹). The mortality percentages were calculated and numbers of each stages resulting from the live treated females were recorded. The symptoms that appear on treated female's mites as a result of death under laboratory conditions were recorded and photographed. The results revealed that, *P. aeruginosa* caused the highest mortality percentage of adult females *T. urticae* 100% after 72h at concentration 10^7 cfu. ml⁻¹ by spraying application. Whereas the mortality percentage were recorded by the dipping technique was 54.58 ± 1.71 . *B. subtilis* also, was recorded increased the average mortality percentage of female *T. urticae* as it reached 73.33 ± 2.67 in spraying technique compared 41.25 ± 2.08 in dipping technique. While in *L. spaericus* recorded 62.08 ± 3.41 mortality (%) in spraying technique but at dipping technique the mortality was 27.50 ± 1.61 . A sharp decrease was shown in number of stages resulting from lived treated females of *T. urticae* with *P. aeruginosa* by four concentrations 10^1 , 10^3 , 10^5 and 10^7 by using spraying technique and the life cy-

cle was stopped at the larval stage only at concentration 10^1 and were not changed into the nymphal stage. While with dipping technique a slight increase in the average number of stages resulting from the adult living females treated with the previously aforementioned concentrations of bacteria *P. aeruginosa*. The present study cleared that application of bacteria strains on *T. urticae* adult females either by spraying or dipping techniques lead to some deformations such as, swelling of the body, then fluids exit followed by complete dryness of the body and forward protrusion of the podosoma region and finally death of the adult females.

Keywords: *Tetranychus urticae*; *Pseudomonas aeruginosa*; *Bacillus subtilis*; *Lysinibacillus spaericus*; Biological control

1 Introduction

The Two-Spotted Spider Mite (TSSM), *Tetranychus urticae* Koch (Acari: Tetranychidae), is a phytophagous mite that is distributed worldwide. This pest pierces the plant epidermis with chelicera and feeds on the cell contents, Decreasing photosynthetic efficiency. (Park and Lee 2002). The spread of the two-spider mite is based on high fecundity of the females, which can lead to a quick population increase (Carey and Bradley 1982). Purpose of

acaricides that the development of pesticide-resistance strains of *T. urticae* and remains the effect problems of these pesticides (Muir and Cranham 1979). A definition of biological control (defined as): “the use of living organisms to decrease injury caused by pests and diseases to possible levels” Biological control methods have become more essential (Fiedler 2012). Biological control factors like predators, parasites, fungi or bacteria can be used for management of mites (Arzanlou et al 2016). Among these organisms, Bacteria, such as *Pseudomonas entomophila*, *Burkholderia* spp., *Chromobacterium* spp., *Streptomyces* spp., *Bacillus* spp. and *Saccharopolyspora* spp., have all recently obtained commercial benefit for production of numerous metabolites that perform as strong insecticides (Ruii 2015). Chitinase enzymes that capable of chitin degradation existent in the cell walls of fungi and insect exoskeletons are thought to be one of the important metabolites produced by these bacteria. Thus, several pathogenic bacteria have a potential role as biological control agents for various fungal and insect pests. (Rathore and Gupta 2015). *Pseudomonas* species have also been shown to be effective as a biological control agent of the two-spotted spider mite pest. (Aksoy et al 2008) indicated that, *Pseudomonas putida*, isolated from tomato plant soils in Turkey, recorded high mortality percentage to the two-spotted spider mites. Furthermore, *Pseudomonas fluorescens* produces bacterial chitinases which are effective to control the mites by hydrolyzing their chitinous exoskeleton (Roobakkumar et al 2011). Strains of *B. thuringiensis* make endotoxins which are lethal to lepidopterous, coleopterous, dipterous insects, mites, nematodes, protozoa and fluke worms (Velooralappil et al 2013). *Paenibacillus polymyxa*, *Bacillus subtilis*, *Bacillus cereus*, *Achromobacter xylosoxidans*, *Pseudomonas aeruginosa* and *Lysinibacillus sphaericus* caused 100, 97.25, 94, 93, 92, 84.29% mortality of *Meloidogyne incognita* after 48 h of treatment. (Soliman et al 2019).

2 Materials and methods

2.1 Rearing of two-spotted spider mite (TSSM), *T. urticae*

2.1.1 Source of *T. urticae*

TSSM was collected from infested tomato and cucumber leaves which are grown in the experimental greenhouse at the campus of Faculty of Agriculture, Ain Shams University, Egypt.

2.2 Mite colony

One newly virgin adult female was transferred by a fine camel hair brush to a sweet potato leaf disc (1mm, in diameter), preserved on a humid cotton wool pad in a Petri dish and left for a period 24-48 h to allow it to lay eggs. The deposited eggs were preserved under laboratory conditions at $25\pm 2^{\circ}\text{C}$, $60\pm 5\%$ R.H. and 16 L: 8 D photoperiod until hatching. The female mite was mounted on a glass slide in Hoyer's media for identification. The recently hatched larvae were transferred singly to sweet potato fresh leaves to follow their developing (Pritam and Clare 1993).

2.3 Bacterial strains

Three rhizobacterial isolates originated from rhizospheric soil collected from different areas at Giza governorate, Egypt. The obtained strains were identified according to 16S rRNA sequence analysis and deposited in the Gen Bank database as *Lysinibacillus sphaericus* (Accession No. LC215050), *Bacillus subtilis* (Accession No. LC215051) and *Pseudomonas aeruginosa* (Accession No. LC215048) were isolated by (Soliman et al 2019).

2.3.1 Growth conditions

Luria Broth medium (LB) was prepared of tryptone 10g, Yeast Extract 5g, NaCl 5gL^{-1} . Strains were grown in complete medium for stored (Peptone 5 g, Yeast extract 3 g, agar 20g and distilled water 1000 ml. (Davis et al 1980).

2.3.2 Preparation of bacterial inoculum

For each bacterial strain, a conical flask (250 ml) containing 100 ml of LB broth medium was inoculated and incubated at 28-30°C with shaking at 150 rpm for 48 h. prior to application. Each one ml distilled water contains 2.50×10^7 colony forming units (cfu. ml⁻¹).

2.4 Bacterial strains as bio-control agent

Mention the names of tested bacteria were evaluated under the laboratory conditions against adult females of *T. urticae*. Twenty healthy adult females per four replicates (5 individuals of female mite/leaf disc) added in petri dish. Four concentrations of bacterial strains were prepared from the main suspension (10^1 , 10^3 , 10^5 , 10^7 cfu. ml⁻¹) in addition to another group of replicates served as a control test. Spraying and dipping application were carried on to each replicate. For spraying technique, after the mites had been placed on the leaf discs, the suspension was sprayed at a distance of 25–30 cm with a hand spray atomizer, replicates served as control were sprayed with distilled water. While for dipping technique, the leaf discs were dipped in each concentration (10^1 , 10^3 , 10^5 , 10^7 cfu. ml⁻¹) for 5 seconds, then *T. urticae* mites were transferred to the treated leaf discs using a fine camel hair brush. Numbers of live and dead individuals were counted after 24, 48 and 72h of all treatments and control. In addition, number of eggs, larvae and nymphs of each concentration for both spraying and dipping application. The percentage mortality of mites was calculated by using the following formula according to (Abbott's 1925).

Recording the numbers of stages resulting from the live treated females at each concentration then recording and photographing the symptoms that appear on the treated females as a result of death due to exposure to bacteria.

2.5 Statistical analysis

Data of all results were analyzed according to Steel and Torrie, 1984. The means were compared by Duncan's Multiple Range Test (DMRT) at 5% clarifying by LSD test (Duncan 1955).

3 Results and Discussion

3.1 Effect of three bacterial strains against *Tetranychus urticae* by using spraying technique.

Data presented in Table 1 showed that, the highest mortality percentage (100%) of adult females *T. urticae* was observed with *P. aeruginosa* after 72h at concentration 10^7 , followed by 10^5 (95%) was recorded, then 10^3 and 10^1 were recorded 90 and 80 (percentage mortality) comparing with the control recorded 15% respectively, while after 24h, the lowest mortality percentage was recorded 55% at concentration 10^1 and the general mean of the dead individuals, 82.50 ± 1.61 was observed compared to that of the control, 11.67 ± 0.93 . In *B. subtilis*, after 72h the highest mortality percentage of adult females was recorded 95% , 85% at concentrations, 10^7 and 10^5 respectively followed by 80% and 75% at concentrations 10^3 and 10^1 respectively but also, after 24h, the lowest mortality percentage was recorded (40%) at concentration 10^1 and general mean of the dead individuals 73.33 ± 2.67 was recorded In *L. spaericus*, also showed high mortality percentage (80%), at concentration 10^7 after 72h followed by 75, 65 and 60% at 10^5 , 10^3 and 10^1 , respectively while after 24h, the lowest mortality percentage (25%) was recorded at concentration 10^1 with a general mean of the dead individuals 62.08 ± 3.41 was recorded in this strain. And so, the results showed highly significant differences among the three bacterial strains. Accordingly, *P. aeruginosa* was more effective on adult females

Table 1. Mortality percentage of adult females *Tetranychus urticae* as affected by three bacterial strains by using spraying technique under laboratory conditions

Type of bacteria	Con. (cfu. ml ⁻¹)	% Mortality After detected hours			General mean± S. E	F. Value	L.S. D
		24h	48 h	72 h			
Control	(water only)	10.00	10.00	15.00	11.67±0.93 d	4.20*	3.22
<i>Pseudomonas aeruginosa</i>	10 ¹	55.00	70.00	80.00	82.50 ±1.61 a	40.20**	5.58
	10 ³	65.00	85.00	90.00			
	10 ⁵	80.00	90.00	95.00			
	10 ⁷	85.00	95.00	100.00			
Mean		71.25 c	85.00 b	91.25 a			
<i>Bacillus subtilis</i>	10 ¹	40.00	65.00	75.00	73.33 ±2.67 b	18.07**	9.23
	10 ³	55.00	70.00	80.00			
	10 ⁵	70.00	75.00	85.00			
	10 ⁷	80.00	90.00	95.00			
Mean		61.25 b	75.00 a	83.75 a			
<i>Lysinibacillus spaericus</i>	10 ¹	25.00	55.00	60.00	62.08 ±3.41 c	8.10*	11.8
	10 ³	45.00	60.00	65.00			
	10 ⁵	65.00	70.00	75.00			
	10 ⁷	70.00	75.00	80.00			
Mean		51.25 b	65.00 a	70 a			

Means with the same letter within each column are not significantly different at the 0.5% level (Duncan's multiple ranges clarifying by LSD test).

of *T. urticae* followed by *B. subtilis* then *L. spaericus* compared to the control and the mortality percentage increase by increasing the concentration and time period. These results are in harmony with that proved by (Mahmoud et al 2020) cleared that, the highest efficiency rate on *T. urticae* was observed with *Acinetobacter* sp. sprays. Mite mortality reached 87.15 and 77.29 % under laboratory and greenhouse conditions respectively after three days of treatment. Also, after seven days of treatment the mortality percentage were significantly lowered 72.22 and 67.11 % and 70.74 and 65.19 % using *B. subtilis* and *B. qassimus* under laboratory and greenhouse conditions. Also, (Li et al 2019) reported that, *Bacillus velezensis* W₁ caused mortality of *T. urticae*, and can be used as a microbial acaricide in the field and the greenhouses. In addition to (Abou Zaid et al 2018) revealed that, when the bacterial strains, *Lysinibacillus sphaericus* and *Bacillus amyloliquefaciens* sprayed on common bean plants to control *T. urticae*, a significant decrease in the population by 37% was observed

after 3 days of the treatment so, it is possible to use these bacterial strains as bio-control agents against *T. urticae*.

Data given in Table 2 revealed that, there was a sharp decrease in the number of the stages resulting from live adult females of *T. urticae* after treatment with the first strain of bacteria *P. aeruginosa* by the four concentrations 10¹, 10³, 10⁵ and 10⁷. Fewer numbers were recorded for egg and larval stages 1.67 and 1.33 individual at the lower concentration 10¹. At 10³, 10⁵ and 10⁷, no numbers were recorded for both stages. Whereas no individuals of nymph stage were recorded in any concentration. Life cycle of *T. urticae* has stopped at the larval stage only at concentration of 10¹ and has not been transformed into the nymph stage. The general average of the moving stages 0.17±0.23 individual was recorded compared to that of the control, and the numbers of eggs, larvae and nymphs were 86, 55 and 66 individuals respectively with general mean of moving stages was 60.50 ±7.72. In the case *B. subtilis*, also, a decrease in numbers of

the moving stages resulting from living adult females after the treatment with the previously aforementioned concentrations was observed. At concentration 10^1 and 10^3 numbers of the eggs and the larvae were 6.00, 3.33 and 2.00, 1.34 individual, respectively. While at the concentrations 10^5 and 10^7 nothing was recorded. The general average of the moving stages 0.42 ± 0.35 individual was recorded, while there were not any nymphs resulted at the all concentrations compared to control. The third tested strain, *L. spaericus* recorded a slight increase in the numbers of the stages compared with (*P. aeruginosa* and *B. subtilis*). The average number of stages recorded for each of the egg, larva and nymph stages at the lowest concentration 10^1 was 8.67, 3.66 and 2.66 individual, respectively. While, in the highest concentration, 10^7 no individuals of any stage were recorded. The data cleared that, *L. spaericus* gave a little effect to some extent when

compared to *P. aeruginosa* and *B. subtilis* due to the appearance of the nymph stage and the general averages 1.45 ± 0.32 individual of moving stages was recorded. The results showed no significant differences between *P. aeruginosa* and *B. subtilis* in the general mean of moving stages comparing with that resulted when *L. spaericus* has applied. These results are in harmony with that proved by (Waked et al 2016). for the spraying technique with the bacterial, *Pseudomonas fluorescens* the highest mortality was recorded 100% and the lowest mortality was 68.75%, 58.75% and 16.25% for the dipping. The longevity was shorter than that of control with both methods, the female longevity averaged 10.32 and 17.04 days with the spraying and the dipping technique while, it was averaged 22.5 and 24.19 days in the control. Generally, the bacterial treatment reduced the number of the deposited eggs to the half when the spraying technique was applied.

Table 2. Average numbers of stages resulting from live adult females of *Tetranychus urticae* as affected by three bacterial strains by using spraying technique under laboratory conditions

Type of bacteria	Mean Number of stages <i>T. urticae</i>				General mean of moving stages \pm S. E	F. Value	L.S. D
	Con. (cfu. ml ⁻¹)	No. of eggs	No. of larvae	No. of nymphs			
Control	(water only)	86.00	55.00	66.00	60.50 ± 7.72 a	0.97	26.71
<i>Pseudomonas aeruginosa</i>	10^1	1.67	1.33	0.00	0.17 ± 0.23 c	1.00**	1.06
	10^3	0.00	0.00	0.00			
	10^5	0.00	0.00	0.00			
	10^7	0.00	0.00	0.00			
<i>Bacillus subtilis</i>	10^1	6.00	2.00	0.00	0.42 ± 0.35 c	2.78**	1.59
	10^3	3.33	1.34	0.00			
	10^5	0.00	0.00	0.00			
	10^7	0.00	0.00	0.00			
<i>Lysinibacillus spaericus</i>	10^1	8.67	3.66	2.66	1.45 ± 0.32 b	1.62*	1.45
	10^3	4.67	2.33	2.00			
	10^5	2.00	1.00	0.00			
	10^7	0.00	0.00	0.00			

Means with the same letter within each column are not significantly different at the 0.5% level (Duncan's multiple ranges clarifying by LSD test).

* = $P < 0.05$, ** = $P < 0.01$

3.2 Effect of three bacterial strains against *Tetranychus urticae* by using the dipping technique

Data given in Table 3 showed that, the highest mortality percentage of adult females, *T. urticae* by using dipping technique, was recorded 80% in case of *P. aeruginosa* after 72h at concentration (10^7). While the lowest mortality percentage was observed at the concentration (10^1) and it was reached 25% after 24h, and the general mean recorded 54.58 ± 1.71 . In *B. subtilis*, the highest mortality percentage of adult females *T. urticae* was recorded 70% at the concentration (10^7) after 72h. While, at 10^1 cfu. ml⁻¹ lowest mortality percentage was recorded (15%) after 24h and the general mean recorded 41.25 ± 2.08 . In *L. spaericus* the lowest concentration (10^1 cfu. ml⁻¹), the mortality percentage reached 5% after 24h, while at the highest concentration (10^7) it reached 50% after 72h. The general mean was 27.50 ± 1.61 compared to that of the control 6.67 ± 0.67 . The results showed highly significant differences between the three bacterial strains. These results resemble those obtained by (Vodovar et al 2006), who compared the mortality of the individuals with both spraying and dipping techniques, there was decreased in the mortality and more outspread after dipping application. This might be attributed to the body surface of *T. urticae* not being moistened and cells of *Pseudomonas putida* biotype B only being insert to the body surface indirectly and at low levels of fertilization, the lower levels of mortality spread over a long time. Also, (Aksoy et al 2008) recorded that, the spraying with the bacteria, *Pseudomonas putida* against *T. urticae*, was more effective, recorded highly significant differences than with the dipping techniques which showed 100% efficiency. Also, resulted in the fewest live eggs.

Data given in Table 4 conducted that, there was a slight increase in the average number of stages resulting from the adult living females after treatment with *P. aeruginosa* by using dipping technique, at the same four concentrations 10^1 , 10^3 , 10^5 and 10^7 . The egg, larva and

nymph stages were only observed and recorded in the lowest concentration 10^1 and they were 5.00, 2.66 and 1.67 individual, respectively. While no stages were observed in the remaining concentrations of 10^3 , 10^5 and 10^7 cfu. ml⁻¹ and the general average for moving stages was 0.54 ± 0.17 individual. In case of *B. subtilis*, there was an increase in the mean number of the eggs, larvae and nymphs with each of the concentrations 10^1 , 10^3 and 10^5 cfu. ml⁻¹. Where at the lowest concentration (10^1 cfu. ml⁻¹), 10.67 individual were recorded, but at the concentration, (10^5 cfu. ml⁻¹) was 3.00 individual for egg stage. While for larva stage it was 5.00 individual at (10^1) concentration, and 1.33 individual at (10^5). As for the nymph stage in the same concentrations (10^1) and (10^5) 4.66 and 1.66 individuals were recorded, while at the highest concentration (10^7) no stages were recorded and the general average of moving stages reached 2.36 ± 0.10 individual compared with *P. aeruginosa*. In case of *L. spaericus*, there was a clear increase in the average number of stages resulting from the treated adult live females for each of the concentrations was observed, whereas, at the lowest concentration (10^1), the average number recorded 19.00, 7.67 and 13.00 individual for the egg, larva and nymph take respectively. While at the highest concentration (10^7) the numbers of stages recorded were 4.33, 1.00 and 2.00 individual respectively, and the general average calculated was 5.17 ± 0.91 individual. For control, recorded 105, 81 and 92 individuals for eggs, larvae and nymphs stage respectively, while the general mean of moving stages recorded was 86.50 ± 2.13 . The results showed highly significant differences between the three bacterial strains in the general mean of moving stages. These results are in harmony with that of (David et al 2019) who revealed that, the application of *B. subtilis* could be considered as an alternative mean to control the two-spotted spider mite under field conditions. Also, (Qessaoui et al 2017) indicated that, the *Pseudomonas fluorescens* isolate Q036B is a promising candidate for biological control of all stages of (TSSM) and has

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Table 3. Mortality percentage of adult females *Tetranychus urticae* as affected by three bacterial strains by using dipping technique under laboratory conditions

Type of bacteria	Con. (cfu. ml ⁻¹)	% Mortality After detected hours			General mean ± S. E	F. Value	L.S. D
		24h	48 h	72 h			
Control	water only	5.00	5.00	10.00	6.67±0.67 d	18.60**	2.32
<i>Pseudomonas aeruginosa</i>	10 ¹	25.00	40.00	55.00	54.58±1.71 a	90.18**	5.94
	10 ³	30.00	55.00	60.00			
	10 ⁵	40.00	70.00	75.00			
	10 ⁷	50.00	75.00	80.00			
Mean		36.25 c	60 b	67.50 a			
<i>Bacillus subtilis</i>	10 ¹	15.00	20.00	35.00	41.25±2.08 b	18.00**	8.16
	10 ³	35.00	40.00	45.00			
	10 ⁵	35.00	45.00	55.00			
	10 ⁷	40.00	60.00	70.00			
Mean		31.25 c	41.25 b	51.25 a			
<i>Lysinibacillus spaericus</i>	10 ¹	5.00	15.00	25.00	27.50±1.61 c	72.00**	4.08
	10 ³	15.00	20.00	35.00			
	10 ⁵	20.00	35.00	40.00			
	10 ⁷	30.00	40.00	50.00			
Mean		17.50 c	27.50 b	37.50 a			

Means with the same letter within each column are not significantly different at the 0.5% level (Duncan's multiple ranges clarifying by LSD test).

* = P < 0.05, ** = P < 0.01

Table 4. Average numbers of stages resulting from live adult females of *Tetranychus. urticae* as affected by three bacterial strains by using dipping technique under laboratory conditions

Type of bacteria	Mean Number of stages of <i>T. urticae</i>				General mean of moving stages ±S.E	F. Value	L.S.D
	Con. (cfu. ml ⁻¹)	No. of eggs	No. of larvae	No. of nymphs			
Control	water only	105.00	81.00	92.00	86.50±2.13 a	18.03**	7.35
<i>Pseudomonas aeruginosa</i>	10 ¹	5.00	2.66	1.67	0.54 ±0.17 d	1.00**	0.79
	10 ³	0.00	0.00	0.00			
	10 ⁵	0.00	0.00	0.00			
	10 ⁷	0.00	0.00	0.00			
<i>Bacillus subtilis</i>	10 ¹	10.67	5.00	4.66	2.36 ±0.10 c	0.13	0.46
	10 ³	5.33	3.20	3.00			
	10 ⁵	3.00	1.33	1.66			
	10 ⁷	0.00	0.00	0.00			
<i>Lysinibacillus spaericus</i>	10 ¹	19.00	7.67	13.00	5.17 ±0.91 b	1.69	4.08
	10 ³	10.00	6.33	5.67			
	10 ⁵	7.33	2.33	3.33			
	10 ⁷	4.33	1.00	2.00			

Means with the same letter within each column are not significantly different at the 0.5% level (Duncan's multiple ranges clarifying by LSD test).

* = P < 0.05, ** = P < 0.01

a potential to contribute to an integrated pest management program to control this important pest.

3.3 Description of the symptoms that appeared on the individuals of adult females of *Tetranychus urticae* after treatment with the three bacterial strains under laboratory conditions

- (A) Adult females of *T. urticae* begin to die after 24 hours, and then a swelling of the body occurred to the body Fig 1.
- (B) Causing an explosion of the contents of the body, and liquid comes out like bloody poisoning
- (C) Dryness of mites occurred and the body's appendages were protruded forward to the podosoma region (Propodosoma + Metapodosoma).
- (D) The body of adult females of *T. urticae* becomes a dry mummy on the leaf surface of the plant.

Results of symptoms that appeared on individuals of adult females after treatment with the tested bacteria were in harmony with that of (Mahmoud et al 2020) who proved that, diagnostic symptoms considered as a common property of the bacterial infection of mites were recorded distinct color changes (dark

brown color with a black tinge), degeneration of internal tissues, discontinuation of feeding, inability to move, weakening of the outer shield, the females failed to oviposit, excretion of diarrhoea-like faeces, occasionally causing mites to turn into glued to the substrate, vomiting and cadavers becoming black due to the bacterial infection. Also, (Wilson et al 2002) reported that, the bacterial hemolysins attacked the blood cell membranes and causes rupturing and body swelling. These hemolysins may also be involved in the pathogenicity of the three bacterial species on *T. urticae*, especially resulted to the rapid mortality. Bacterial pathogens invade their hosts through the mouth and the alimentary canal. Less often, they enter through peritremes and wounds in the integument (Poinar and Poinar 1998). He showed that, the bacterial infections perhaps classified as toxemia, when bacteria are confined in the alimentary canal where they produce toxins causes septicaemia and when bacteria multiply in the hemocoel, and the mites will be killed (Tanada and Kaya 1993) and (Raaijmakers et al 2002), they indicated that, pathogenic bacteria *Pseudomonas* affect the cell surface of the host, this adhesion of bacteria may increase the entry of proteases, chitinases and lipases through the epidermis, stigmata and body openings of *T. urticae*, which can lead to rapidly death of the mites.

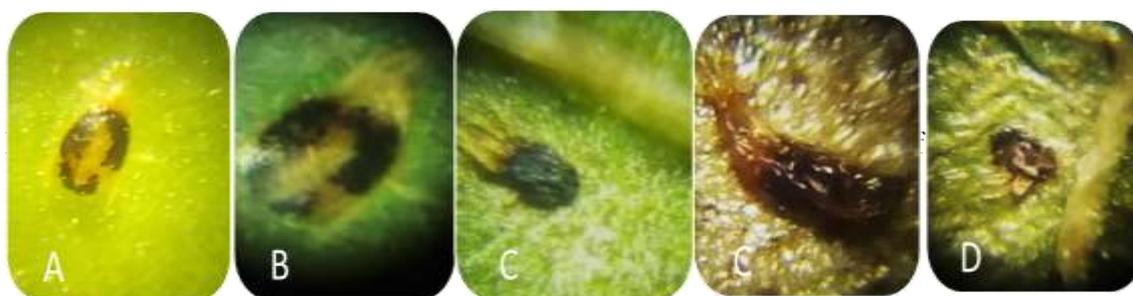


Fig 1. Description of the symptoms that appeared on individuals of adult females of *Tetranychus. urticae* after treatment with the three bacterial strains

4 Conclusion

The mentioned scientific names of the bacteria cleared high mortality percentage varying according to concentration and according to the method of application, either through spraying or dipping techniques, the spraying technique revealed a significant difference and increase in the mortality percentage compared to that through the dipping technique, *P. aeruginosa* was the most effective at the concentration (10^7 cfu. ml⁻¹), using the spraying technique. Also, as a result of the exposure to bacteria there were symptoms of death beginning with swelling of the body and ended with complete dryness have been evolved.

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كفاءة ثلاثة أنواع من السلالات البكتيرية ضد *Tetranychus urticae* Koch تحت الظروف المعملية (Acari: Tetranychidae)

[31]

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B. subtilis فقد تم تسجيل أعلى متوسط لنسبة الموت لإناث الأكاروس 2.67 ± 73.33 في طريقة الرش مقارنة بطريقة الغمر 2.08 ± 41.25 . بينما في السلالة *L. spaericus* سجل متوسط نسبة الموت في طريقة الرش 3.41 ± 62.08 لكن سجلات في طريقة الغمر 1.16 ± 27.50 . أيضا حدث إنخفاض حاد في عدد الأطوار الناتجة من الإناث الحية لأكاروس *T. urticae* عند استخدام *P. aeruginosa* بالاربعة تركيزات $10^1, 10^3, 10^5, 10^7$ وذلك باستخدام طريقة الرش، ولوحظ أيضا توقف دورة الحياة عند طور اليرقة فقط عند تركيز 10^1 ولم تتحول إلي طور الحورية. بينما في طريقة الغمر لوحظ زيادة طفيفة في متوسط أعداد الأطوار الناتجة من الإناث الحية بعد المعاملة أيضا بنوع البكتيريا الأول *P. aeruginosa* وبنفس التركيزات. وفي النهاية كنتيجة للتعرض للبكتيريا سواء بطريقة الرش أو الغمر ظهور أعراض نتيجة الموت وتتمثل في إنتفاخ في الجسم ثم خروج سائل يتبعه جفاف كامل للجسم وبروز الأطراف للأمام لكل من منطقة الأرجل الأمامية والخلفية.

الموجز

تهدف هذه الدراسة إلي تقييم كفاءة ثلاثة أنواع من السلالات البكتيرية وهي *Bacillus subtilis*، *Pseudomonas aeruginosa* و *Lysinibacillus spaericus* ضد الإناث الكاملة لأكاروس العنكبوت الأحمر ذي البقعتين *Tetranychus urticae* وذلك باستخدام طريقة الرش والغمر بتركيزات $10^1, 10^3, 10^5, 10^7$ (عدد الخلايا البكتيرية في 1 مل معلق)، وكذلك حساب نسب الموت لأعداد الأطوار الناتجة من الإناث الحية المعاملة، هذا بالإضافة إلي تصوير وتسجيل الأعراض التي ظهرت علي إناث الأكاروس المعاملة بالبكتيريا نتيجة للموت وذلك تحت الظروف المعملية. وقد أشارت النتائج إلي تسجيل أعلى نسبة موت للإناث الكاملة من الأكاروس وصلت إلي 100% في طريقة الرش بنوع البكتيريا *P. aeruginosa* بعد 72 ساعة بتركيز 10^7 cfu (10^7 cfu) في حين أنه تم تسجيل نسبة موت $54.58 \pm$ باستخدام طريقة الغمر. أما في السلالة