



# *Taifor Induced Changes in the Growth and Plant Growth Promoting Traits of Bacteria Isolated from Tomato Crop Field*

R. Shanmugavalli and S. Umamaheswari

Research Journal of Agricultural Sciences  
An International Journal

P- ISSN: 0976-1675

E- ISSN: 2249-4538

Volume: 13

Issue: 03

*Res. Jr. of Agril. Sci. (2022) 13: 653–658*



# Tafgor Induced Changes in the Growth and Plant Growth Promoting Traits of Bacteria Isolated from Tomato Crop Field

R. Shanmugavalli<sup>1</sup> and S. Umamaheswari<sup>\*2</sup>

Received: 22 Nov 2021 | Revised accepted: 02 May 2022 | Published online: 24 May 2022

© CARAS (Centre for Advanced Research in Agricultural Sciences) 2022

## ABSTRACT

The persistence of tafgor pesticide and their residues in tomato crop fields could cause adverse impact on the non-target organisms. With this view, the present study was initiated to observe the impact of tafgor on the bacteria prevalent in tomato crop field soil. Bacteria are important component in the soil ecosystem as they play a vital role in nutrient cycling and promote the growth of plants. In this study, the effect of Tafgor pesticide on the growth of *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca* and their plant growth promoting traits like indole-3-acetic acid (IAA) production and siderophore production were assayed. The findings of this study reveal that tafgor significantly decreased the bacterial growth and consequently reduced the IAA and siderophore production. Thus, the usage of tafgor could alter the bacterial diversity and interfere with the nutrient cycle and plant growth, which may affect the crop yield and quality.

**Key words:** Tafgor, *Bacillus sp.*, *Pseudomonas polymyca*, Indole-3-acetic acid (IAA), Siderophore

The minimization of pesticide impact on non-target organisms like bacteria, fungi, insects are of prime concern. Only 5% of the pesticides applied reach the target pest, rest of it contaminates the soil and water bodies. The constant leaching of these pesticide residues contaminates the environment, causing multifaceted toxicity to the biota [1]. Tafgor is a systemic organophosphorus insecticide applied in tomato, onion, chilli, cauliflower, okra, mango, brinjal, cabbage crop fields to kill sucking and caterpillar pest bug, stem borer, shoot fly, beetles, thrips, aphids, mites etc., long term application of dichloropropane-dichloropropene, fosfiazate and chloropicrin reduces the bacterial biomass in the agricultural field [2]. Bacteria are known to produce phytohormones, which play an important role in various physiological processes [3]. Several bacteria like (*Pseudomonas fragi*, *Bacillus cereus*, *Rhizobium*, *Bacillus aerius*, *Bacillus amyloliquefaciens*, *Azotobacter*, fluorescent *Pseudomonas*, *Mesorhizobium*, *Azospirillum brasilense*, *Serratia spp.*, *Enterobacter sp.*, *Bacillus subtilis*, *Methylobacterium* genus, *Pseudomonas fluorescens*, *P. chlororaphis*, *Sphingomonas sp.* produce siderophore and IAA [4-12]. Keeping in this view, the present study was initiated to study the impact of Tafgor pesticide on the growth of tomato crop field soil bacteria and to assay the production of IAA and siderophore.

## MATERIALS AND METHODS

### Determination of growth curve of bacteria in minimal salt medium under Tafgor pesticide stress

100 µl ( $21 \times 10^9$  cfu / ml) of 48 hours of *Bacillus licheniformis* nutrient broth culture was inoculated into conical flask containing 100 ml of sterilised minimal salt medium and Tafgor pesticide was added (15 ppm and 30 ppm) separately and kept in the orbital shaker at 150 rpm at 30°C for 96 hours in triplicates. Simultaneously, control was maintained without addition of pesticide in minimal salt medium containing bacteria. 3 ml of sample was withdrawn and the optical density at initial, 4<sup>th</sup>, 8<sup>th</sup>, 16<sup>th</sup>, 24<sup>th</sup>, 48<sup>th</sup>, 72<sup>nd</sup> and 96 hours OD was read at 620 nm using colorimeter. Simultaneously, colony forming units on nutrient agar plates of each test and bacterial control were recorded from 100 µl of  $10^{-9}$  dilution factor. Same procedure was followed for *Bacillus cereus* and *Paenibacillus polymyca*.

### Impact of Tafgor pesticide on IAA production by *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca*

Indole-3-acetic acid synthesized by bacterial strains was quantitatively evaluated by the method of Gordon and Weber [13] and later modified by Brick *et al.* [14]. Selected bacteria strains were grown in Luria Bertani (LB) broth. Luria Bertani broth (100 ml) having fixed concentration of tryptophan (100 mg/ml) and supplemented with 15 ppm and 30 ppm of recommended rate of each pesticide was inoculated with 1 ml culture of *Bacillus licheniformis* bacterial isolates (cfu/ml) and was incubated for seven days at  $28 \pm 2^\circ\text{C}$  with shaking at 120 rpm. Simultaneously, control was maintained without addition of pesticide containing bacteria. After seven days, 5 ml of

\* S. Umamaheswari

✉ umadurai73@yahoo.com

<sup>1-2</sup> Department of Zoology, Periyar EVR College (Affiliated to Bharathidasan University), Tiruchirappalli - 620 023, Tamil Nadu, India

culture of each treatment was centrifuged (9,000 rpm) for 15 minutes and an aliquot of 2 ml supernatant was mixed 100  $\mu$ l of orthophosphoric acid and 4 ml of salkowsky reagent (2% 0.5 M  $\text{FeCl}_3$  in 35% per-chloric acid) and incubated at  $28 \pm 2^\circ\text{C}$  in darkness for 1 hour. The absorbance of developed pink colour was read at 530 nm. IAA concentration in the supernatant was determined using a calibration curve of pure IAA as a standard. Cfu also was determined simultaneously. Same procedure was followed for *Bacillus cereus* and *Paenibacillus polymyca*.

#### Impact of Taigor pesticide on siderophore production by *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca* [15]

Siderophore production was studied by inoculating *Bacillus licheniformis* in conical flask containing 1200 ml succinate medium ( $\text{K}_2\text{HPO}_4$  6.0,  $\text{KH}_2\text{PO}_4$  3.0,  $\text{MgSO}_4$  0.2,  $(\text{NH}_4)_2\text{SO}_4$  1.0 and succinic acid 4.0 gm  $\text{L}^{-1}$ , pH 7.0) and supplemented with 15 ppm and 30 ppm of Taigor pesticide. *Bacillus licheniformis* was inoculated and incubated in succinate medium for 24 – 72 hours at  $28^\circ\text{C}$  with constant shaking at 120 rpm on rotary shaker. Simultaneously, control was maintained without addition of pesticide. After incubation, the fermented broth were centrifuged at 10,000 rpm in cooling centrifuge at  $4^\circ\text{C}$  for 10 minutes and cell free supernatant was then mixed with 0.5 ml CAS (Chrome Azurol Solution) solution

and 10  $\mu$ l shuttling solution (sulfosalicyclic acid). The optical density was read in the spectrophotometer at absorbance 630 nm. The percentage of siderophore units was estimated as the proportion of CAS color shifted using the formula  $[(\text{Ar}-\text{As}) / \text{Ar}] \times 100$ , where Ar is the A630 nm of reference sample (medium + CAS assay solution + shuttle solution) and As is the A630 nm of the sample (supernatant + CAS assay solution + shuttle solution). Colony forming units (cfu) also was determined simultaneously. Same procedure was followed for *Bacillus cereus* and *Paenibacillus polymyca*.

#### Statistical analysis

The data obtained were subjected to one way and two-way ANOVA using version SPSS 16.0.

## RESULTS AND DISCUSSION

The growth curve of bacteria displayed in (Fig 1-9), reveal that the lag phase, log phase and decline phase lasted till 16 hours, 56 hours and 24 hours in terms of optical density (OD) respectively, irrespective of the species and taigor pesticide concentration. Dose-dependent decline in bacterial population in terms of optical density (Table 1-2) and colony forming units (cfu) (Table 3-4) was evinced in bacteria exposed to Taigor, irrespective of the species.

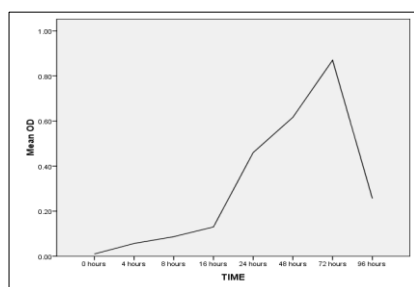


Fig 1 Growth curve (OD) of *Bacillus licheniformis* in minimal salt medium control

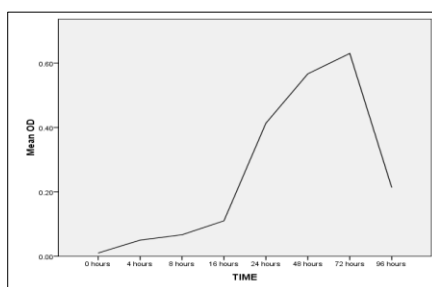


Fig 2 Growth curve (OD) of *Bacillus licheniformis* in minimal salt medium containing 15 ppm of Taigor pesticide

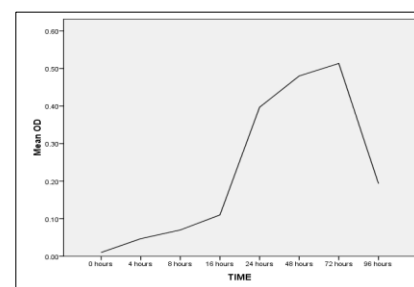


Fig 3 Growth curve (OD) of *Bacillus licheniformis* in minimal salt medium containing 30 ppm of Taigor pesticide

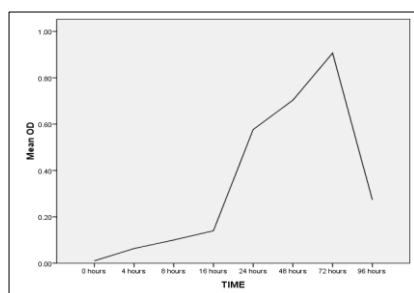


Fig 4 Growth curve (OD) of *Bacillus cereus* in minimal salt medium control

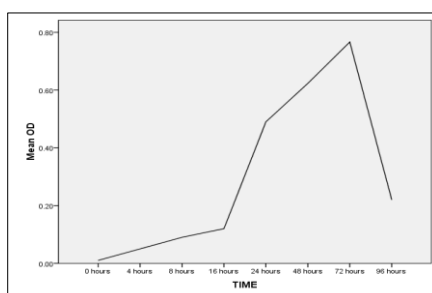


Fig 5 Growth curve (OD) of *Bacillus cereus* in minimal salt medium containing 15 ppm of Taigor pesticide

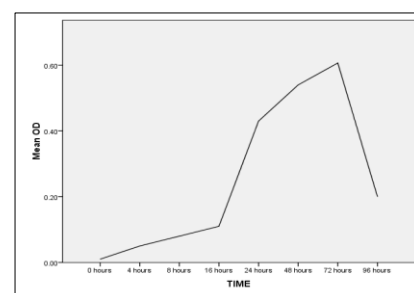


Fig 6 Growth curve (OD) of *Bacillus cereus* in minimal salt medium containing 30 ppm of Taigor pesticide

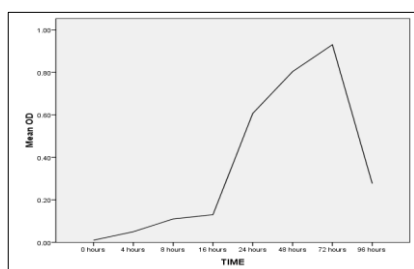


Fig 7 Growth curve (OD) of *Paenibacillus polymyca* in minimal salt medium control

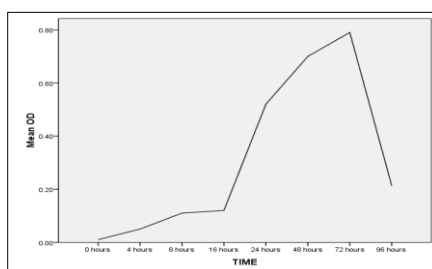


Fig 8 Growth curve (OD) of *Paenibacillus polymyca* in minimal salt medium containing 15 ppm of Taigor pesticide

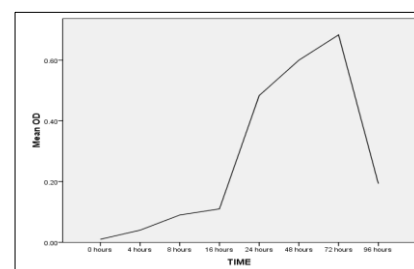


Fig 9. Growth curve (OD) of *Paenibacillus polymyca* in minimal salt medium containing 30 ppm of Taigor pesticide

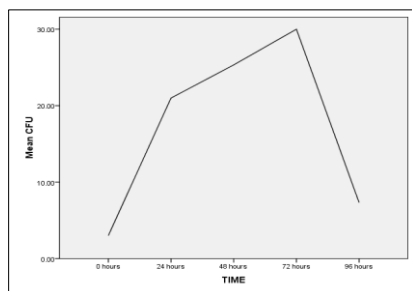


Fig 10 Growth curve of *Bacillus licheniformis* in minimal salt medium control ( $\times 10^{-9}$  cfu /ml)

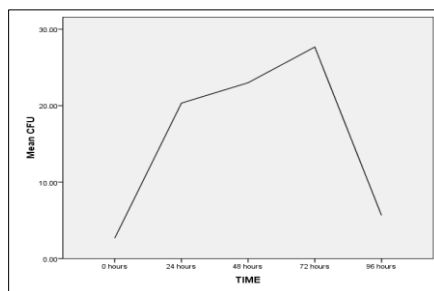


Fig 11 Growth curve of *Bacillus licheniformis* in minimal salt medium with Tafgor pesticide (15 ppm) ( $\times 10^{-9}$  cfu /ml)

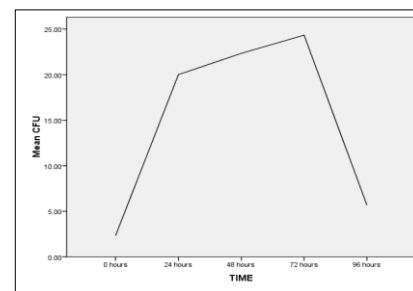


Fig 12 Growth curve of *Bacillus licheniformis* in minimal salt medium with Tafgor pesticide (30 ppm) ( $\times 10^{-9}$  cfu /ml)

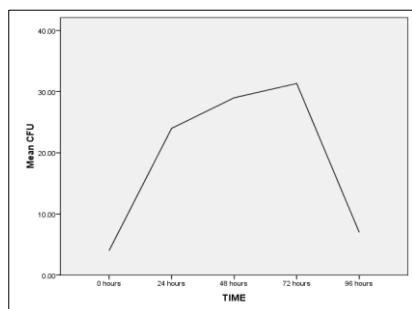


Fig 13 Growth curve of *Bacillus cereus* in minimal salt medium control ( $\times 10^{-9}$  cfu /ml)

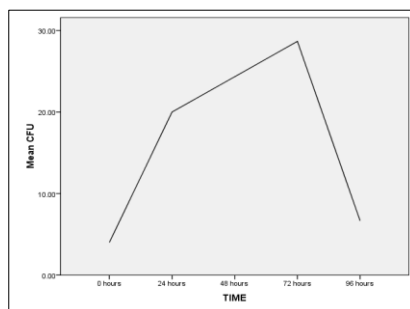


Fig 14 Growth curve of *Bacillus cereus* in minimal salt medium with Tafgor pesticide (15 ppm) ( $\times 10^{-9}$  cfu /ml)

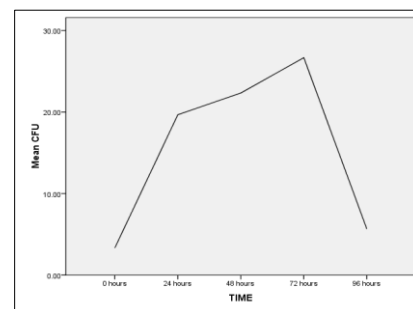


Fig 15 Growth curve of *Bacillus cereus* in minimal salt medium with Tafgor pesticide (30 ppm) ( $\times 10^{-9}$  cfu /ml)

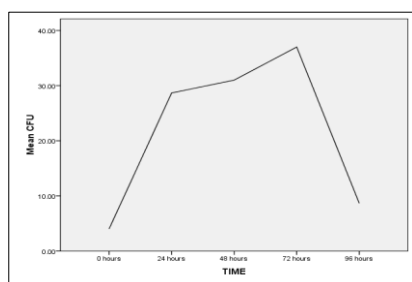


Fig 16 Growth curve of *Paenibacillus polymyca* in minimal salt medium control ( $\times 10^{-9}$  cfu /ml)

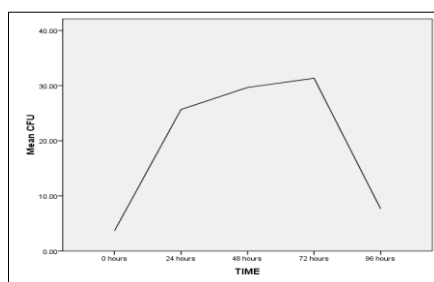


Fig 17 Growth curve of *Paenibacillus polymyca* in minimal salt medium with Tafgor pesticide (15 ppm) ( $\times 10^{-9}$  cfu /ml)

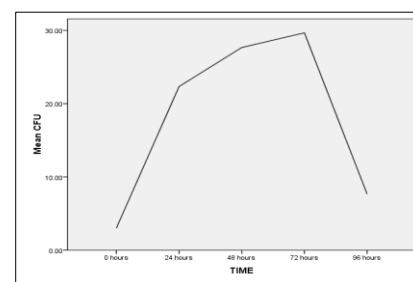


Fig 18 Growth curve of *Paenibacillus polymyca* in minimal salt medium with Tafgor pesticide (30 ppm) ( $\times 10^{-9}$  cfu /ml)

Table 1 Variation in the growth (OD) of bacteria on exposure to Tafgor pesticide in minimal salt medium

Treatment	<i>Bacillus licheniformis</i>	<i>Bacillus cereus</i>	<i>Paenibacillus polymyca</i>
Control	0.3108 <sup>a</sup>	0.3467 <sup>a</sup>	0.3646 <sup>a</sup>
15 ppm	0.2575 <sup>b</sup>	0.2962 <sup>b</sup>	0.3142 <sup>b</sup>
30 ppm	0.2275 <sup>c</sup>	0.2533 <sup>c</sup>	0.2763 <sup>c</sup>
F value	1.710E3 <sup>***</sup>	1.886E3 <sup>***</sup>	4.848E3 <sup>***</sup>
P	0.001	0.001	0.001

n = 3, Values are mean, \*\*\*Significant at  $P < 0.001$ , In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Table 2 Growth of bacteria on exposure to Tafgor pesticide in minimal salt medium

Time	<i>Bacillus licheniformis</i>	<i>Bacillus cereus</i>	<i>Paenibacillus polymyca</i>
0 hours	0.0100 <sup>h</sup>	0.0100 <sup>h</sup>	0.0100 <sup>h</sup>
4 hours	0.0511 <sup>g</sup>	0.0544 <sup>g</sup>	0.0467 <sup>g</sup>
8 hours	0.0744 <sup>f</sup>	0.0900 <sup>f</sup>	0.1033 <sup>f</sup>
16 hours	0.1167 <sup>e</sup>	0.1233 <sup>e</sup>	0.1200 <sup>e</sup>
24 hours	0.4233 <sup>c</sup>	0.4989 <sup>c</sup>	0.5367 <sup>c</sup>
48 hours	0.5544 <sup>b</sup>	0.6222 <sup>b</sup>	0.7011 <sup>b</sup>
72 hours	0.6711 <sup>a</sup>	0.7600 <sup>a</sup>	0.8011 <sup>a</sup>
96 hours	0.2211 <sup>d</sup>	0.2311 <sup>d</sup>	0.2278 <sup>d</sup>

F value	2.287E4 <sup>***</sup>	2.682E4 <sup>***</sup>	9.124E4 <sup>***</sup>
P	0.001	0.001	0.001

n = 3, Values are mean, \*\*\*Significant at  $P < 0.001$ , In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Table 3 Variation in the growth (cfu) of bacteria on exposure to Tafgor pesticide in minimal salt medium

Treatment ( $\times 10^9$ cfu/ml)	<i>Bacillus licheniformis</i> ( $\times 10^9$ cfu/ml)	<i>Bacillus cereus</i> ( $\times 10^9$ cfu/ml)	<i>Paenibacillus polymyca</i> ( $\times 10^9$ cfu/ml)
Control	17.3333 <sup>a</sup>	19.0667 <sup>a</sup>	21.8667 <sup>a</sup>
15 ppm	15.8667 <sup>b</sup>	16.7333 <sup>b</sup>	19.6000 <sup>b</sup>
30 ppm	15.0000 <sup>c</sup>	15.5333 <sup>c</sup>	18.0667 <sup>c</sup>
F value	58.688 <sup>***</sup>	242.111 <sup>***</sup>	176.214 <sup>***</sup>
P value	0.001	0.001	0.001

cfu- Colony forming units, \*\*\*Significant at  $P < 0.001$ , n = 3, Values are expressed as mean, In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Irrespective of the bacterial species and tafgor concentration, the growth curve of bacteria depicted in (Fig 10-18, Table 3-4) exhibited similar growth pattern in terms of cfu

(lag phase: 24 hours; log phase: 48 hours and decline phase: 24 hours).

Table 4 Variation in the population of bacteria exposure to Tafgor pesticide in minimal salt medium

Time	<i>Bacillus licheniformis</i> ( $\times 10^9$ cfu/ml)	<i>Bacillus cereus</i> ( $\times 10^9$ cfu/ml)	<i>Paenibacillus polymyca</i> ( $\times 10^9$ cfu/ml)
0 hours	2.7778 <sup>c</sup>	3.7778 <sup>c</sup>	3.5556 <sup>c</sup>
24 hours	20.4444 <sup>c</sup>	21.2222 <sup>c</sup>	25.5556 <sup>c</sup>
48 hours	23.5556 <sup>b</sup>	25.2222 <sup>b</sup>	29.4444 <sup>b</sup>
72 hours	27.3333 <sup>a</sup>	28.8889 <sup>a</sup>	32.6667 <sup>a</sup>
96 hours	6.2222 <sup>d</sup>	6.4444 <sup>d</sup>	8.0000 <sup>d</sup>
F value	3.010E3 <sup>***</sup>	5.771E3 <sup>***</sup>	5.025E3 <sup>***</sup>
P	0.001	0.001	0.001

cfu- Colony forming units, \*\*\*Significant at  $P < 0.001$ ,  $n = 3$ , Values are expressed as mean, In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Table 5 Impact of Tafgor pesticide IAA production by *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca*

Treatment	Indole-3-Acetic Acid ( $\mu$ l/ml)		
	<i>Bacillus licheniformis</i>	<i>Bacillus cereus</i>	<i>Paenibacillus polymyca</i>
Control	0.7033 $\pm$ 0.00333 <sup>a</sup>	0.7333 $\pm$ 0.00333 <sup>a</sup>	0.8867 $\pm$ 0.00333 <sup>a</sup>
15 ppm	0.4867 $\pm$ 0.00333 <sup>b</sup>	0.5033 $\pm$ 0.00333 <sup>b</sup>	0.4067 $\pm$ 0.00333 <sup>b</sup>
30 ppm	0.2833 $\pm$ 0.00333 <sup>c</sup>	0.3050 $\pm$ 0.00500 <sup>c</sup>	0.2633 $\pm$ 0.00333 <sup>c</sup>
F value	3.970E3 <sup>***</sup>	3.090E3 <sup>***</sup>	9.592E3 <sup>***</sup>
P	0.001	0.001	0.001

$n = 3$ , values are mean  $\pm$  Standard Error, \*\*\*Significant at  $P < 0.001$ , In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Table 6 *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca* population on the 7 days of Tafgor pesticide exposure of IAA production

Treatment ( $\times 10^9$ cfu/ml)	<i>Bacillus licheniformis</i> ( $\times 10^9$ cfu/ml)	<i>Bacillus cereus</i> ( $\times 10^9$ cfu/ml)	<i>Paenibacillus polymyca</i> ( $\times 10^9$ cfu/ml)
Control	17.3333 $\pm$ 0.33333 <sup>a</sup>	19.3333 $\pm$ 0.33333 <sup>a</sup>	18.6667 $\pm$ 0.33333 <sup>a</sup>
15 ppm	11.6667 $\pm$ 0.33333 <sup>b</sup>	13.3333 $\pm$ 0.33333 <sup>b</sup>	10.6667 $\pm$ 0.33333 <sup>b</sup>
30 ppm	10.3333 $\pm$ 0.33333 <sup>c</sup>	12.3333 $\pm$ 0.33333 <sup>c</sup>	9.3333 $\pm$ 0.33333 <sup>c</sup>
F value	124.333 <sup>***</sup>	129.000 <sup>***</sup>	229.333 <sup>***</sup>
P value	0.001	0.001	0.001

cfu- Colony forming units, \*\*\* Significant at  $P < 0.001$ ,  $n = 3$ , Values are expressed as mean  $\pm$  Standard Error, In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

From the (Table 5), it is evident that IAA produced by *Bacillus licheniformis* significantly ( $F = 3.970E3$ ;  $P < 0.001$ ) declined on exposure to tafgor (control:  $0.7033 \pm 0.00333$ ; 15 ppm:  $0.4867 \pm 0.00333$ ; 30 ppm:  $0.2833 \pm 0.00333$ ). Production of IAA by *Bacillus cereus* significantly ( $F = 3.090E3$ ;  $P < 0.001$ ; control:  $0.7333 \pm 0.00333$ ; 15 ppm:  $0.5033 \pm 0.00333$ ; 30 ppm:  $0.3050 \pm 0.00500$ ) reduced on exposure to tafgor. Similar response was evinced with respect to *Paenibacillus polymyca* (control:  $0.8867 \pm 0.00333$ ; 15 ppm:  $0.4067 \pm$

$0.00333$ ; 30 ppm:  $0.2633 \pm 0.00333$ ;  $F = 9.592E3$ ;  $P < 0.001$ ). Simultaneously, significant ( $F = 124.333$ ,  $P < 0.001$ ) decline in the *Bacillus licheniformis* population was evinced during IAA production (control:  $17.3333 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $11.6667 \pm 0.33333 \times 10^9$  cfu / ml; 30 ppm:  $10.3333 \pm 0.33333 \times 10^9$  cfu / ml). Similar response was elicited by *Bacillus cereus* (control:  $19.3333 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $13.3333 \pm 0.33333 \times 10^9$  cfu / ml; 30 ppm:  $12.3333 \pm 0.33333 \times 10^9$  cfu / ml;  $F = 129.000$ ;  $P < 0.001$ ) and *Paenibacillus polymyca* (control:  $18.6667 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $10.6667 \pm 0.33333 \times 10^9$  cfu / ml; 30 ppm:  $9.3333 \pm 0.33333 \times 10^9$  cfu / ml;  $F = 229.333$ ;  $P < 0.001$ ) (Table 6).

Table 7 Impact of Tafgor pesticide Siderophore production by *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca*

Treatment	Siderophore production (%)		
	<i>Bacillus licheniformis</i>	<i>Bacillus cereus</i>	<i>Paenibacillus polymyca</i>
Control	0.8133 $\pm$ 0.00333 <sup>a</sup>	0.8633 $\pm$ 0.00333 <sup>a</sup>	0.8767 $\pm$ 0.00333 <sup>a</sup>
15 ppm	0.4467 $\pm$ 0.00333 <sup>b</sup>	0.5267 $\pm$ 0.00333 <sup>b</sup>	0.4367 $\pm$ 0.00333 <sup>b</sup>
30 ppm	0.3367 $\pm$ 0.00333 <sup>c</sup>	0.4167 $\pm$ 0.00667 <sup>c</sup>	0.3167 $\pm$ 0.00333 <sup>c</sup>
F value	5.606E3 <sup>***</sup>	2.437E3 <sup>***</sup>	7.824E3
P	0.001	0.001	0.001

$n = 3$ , values are mean  $\pm$  Standard Error, \*\*\*Significant at  $P < 0.001$ , In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

Table 8 *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca* population on the 7 days of Tafgor pesticide exposure of Siderophore production

Treatment ( $\times 10^9$ cfu/ml)	<i>Bacillus licheniformis</i> ( $\times 10^9$ cfu/ml)	<i>Bacillus cereus</i> ( $\times 10^9$ cfu/ml)	<i>Paenibacillus polymyca</i> ( $\times 10^9$ cfu/ml)
Control	10.6667 $\pm$ 0.33333 <sup>a</sup>	11.3333 $\pm$ 0.33333 <sup>a</sup>	10.3333 $\pm$ 0.33333 <sup>a</sup>
15 ppm	8.6667 $\pm$ 0.33333 <sup>b</sup>	10.3333 $\pm$ 0.33333 <sup>b</sup>	7.6667 $\pm$ 0.33333 <sup>b</sup>
30 ppm	7.3333 $\pm$ 0.33333 <sup>c</sup>	8.6667 $\pm$ 0.33333 <sup>c</sup>	6.3333 $\pm$ 0.33333 <sup>c</sup>
F value	25.333 <sup>***</sup>	16.333 <sup>**</sup>	37.333 <sup>***</sup>
P value	0.001	0.01	0.001

cfu- Colony forming units, \*\*\* Significant at  $P < 0.001$  and  $P < 0.01$ ,  $n = 3$ , Values are expressed as mean  $\pm$  Standard Error, In a column, figures having dissimilar letters differ significantly according to Duncan New Multiple Range Test (DMRT)

On exposure to tafgor pesticide, bacterial production of siderophore significantly declined, irrespective of all species (*Bacillus licheniformis*: control:  $0.8133 \pm 0.00333$ ; 15 ppm:  $0.4467 \pm 0.00333$ ; 30 ppm:  $0.3367 \pm 0.00333$ ;  $F = 5.606E3$ ;  $P < 0.001$ , *Bacillus cereus*: control:  $0.8633 \pm 0.00333$ ; 15 ppm:  $0.5267 \pm 0.00333$ ; 30 ppm:  $0.4167 \pm 0.00667$ ;  $F = 2.437E3$ ;  $P < 0.001$ , *Paenibacillus polymyca*: control:  $0.8767 \pm 0.00333$ ; 15 ppm:  $0.4367 \pm 0.00333$ ; 30 ppm:  $0.3167 \pm 0.00333$ ;  $F = 7.824E3$ ;  $P < 0.001$ ). Moreover, the decrease in siderophore production was found to be dose-dependent. As the concentration of Tafgor increased, production of siderophore by bacteria decreased (Table 7). Furthermore, the population of Tafgor pesticide exposed bacteria significantly reduced during siderophore production, which could be the possible cause for significantly reduction in siderophore. (*Bacillus licheniformis*: control:  $10.6667 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $8.6667 \pm$



$0.33333 \times 10^9$  cfu / ml; 30 ppm:  $7.3333 \pm 0.33333 \times 10^9$  cfu / ml; F = 25.333; P < 0.001, *Bacillus cereus*: control:  $11.3333 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $10.3333 \pm 0.33333 \times 10^9$  cfu / ml; 30 ppm:  $8.6667 \pm 0.33333 \times 10^9$  cfu / ml; F = 16.333; P < 0.01, *Paenibacillus polymyca*: control:  $10.3333 \pm 0.33333 \times 10^9$  cfu / ml; 15 ppm:  $7.6667 \pm 0.33333 \times 10^9$  cfu / ml; 30 ppm:  $6.3333 \pm 0.33333 \times 10^9$  cfu / ml; F = 37.333; P < 0.001) (Table 8).

Dose dependent inhibition of bacterial population observed in this study is in agreement with the findings of Shetti and Kaliwal [16], who have also observed similar dose-dependent inhibitory effect on Imidacloprid on soil bacteria both under laboratory and field conditions. Further, they have also evinced that the toxic effect of Imidacloprid disappeared after a period of 28 days of application in the field. In agreement with the present findings, Shetti *et al.* [17] have also reported dose-dependent inhibition of growth of *Escherichia coli* due to Imidacloprid exposure in minimal salt medium. Imidacloprid induced toxicity in aquatic bacteria *Vibrio fischeri* [18]. Shetti and Kaliwal [19] have reported significant decrease in the growth of *Bacillus weihenstephanensis* exposed to imidacloprid ( $10^{-7}$  to  $10^{-3}$  M) for a period of 96 hours in terms of optical density. In consistent with the present findings, Ahemad and Khan [20] have also evinced that pesticides (metribuzin, glyphosate, imidacloprid, thiamethoxam, hexaconazole, metalaxyl and kitazin) elicited dose-dependent decline in the production of siderophore, IAA, 2,3-dihydroxy benzoic acid and exo-polysaccharides by *Mesorhizobium sp.* As evinced in this study, Mohite [21] have also reported that *Bacillus megaterium*, *Lactobacillus acidophilus*, *Lactobacillus casei*, *Bacillus cereus* and *Bacillus subtilis* produce IAA.

Similarly, Ali *et al.* [22] have also observed that the bacterial strain *Bacillus*, *Pseudomonas*, *Escherichia*, *Micrococcus*, *Staphylococcus* genera produce IAA. In the opinion of Patten and Glick [23], the role of bacterial IAA is to stimulate plant growth and suppress diseases. Several growth and environmental conditions do influence the production of IAA by bacteria [24]. The present finding is in good accord with the observations of Ghosh *et al.* [25] who have also reported that bacteria like *Bacillus subtilis*, *Bacillus megatericus* and *Pseudomonas aerogenosa* produce siderophore. Siderophore producing bacteria (*E. coli*, *Pseudomonas fluorescence*, *Rhizopus sp.* and *Aspergillus flavus*) have been reported by Kannahi and Senbagam [26].

As observed in this study, Kumar *et al.* [27] have isolated siderophore producing *Bacillus sp* and *Enterobacter sp.* from soil. Similarly, Ahemad and Khan [28] have reported that adverse impact of pesticides on *Pseudomonas putida* strain isolated from rhizosphere soil samples of *Brassica campestris*.

As the concentration of pesticides increased (fipronil, pyriproxyfen, imidacloprid and thiamethoxam) there was progressive decrease in the production of siderophore and IAA. In agreement with the findings, Chennappa *et al.* [29] have reported that production of IAA by *Azotobacter vinelandii*, *A. tropicalis*, *A. armeniacus* and *A. salinestrus* isolated from paddy crop field soil was reduced due to pendimethalin, chloropyrifos, glyphosate and phorate exposure. Present findings collaborate with the observation of Aswathi *et al.* [30] who have evinced that *Pseudomonas fluorescens* evinced maximum tolerance to imidacloprid at lower concentration of imidacloprid (0.01%), pendimethalin (0.2%) and carbendazim (0.05%). Furthermore, *Pseudomonas fluorescens* produced siderophore and IAA.

The results are in confirmity with Panomkhum *et al.* [31] have reported that application of thiamethoxam in the Cassava cultivar Rayong decreased the bacterial population. On contrary, they have reported that the bacteria exhibited the ability to produce IAA and siderophore even in the presence of thiamethoxam. IAA and siderophore production by bacteria enhanced in the presence of organic fertilizer. Similar observations have been evinced by Tripti *et al.* [32] who have reported that pesticides reduced IAA production by *Burkholderia sp.* isolated from agricultural field soil at Dhanbad region, Jharkhand in a dose-dependent manner. The report of Swarupa and Kumar [33] were also similar, who have demonstrated that bacteria could use Chlorpyrifos as a sole source of carbon. The results are in accordance with those of Kumar [34] who have observed dose-dependent decline in siderophore production by *Pseudomonas fluorescens* as exposure to 25 and 200 ppm of pesticides like acephate, glyphosate, monocrotophos and phorate. The results obtained reveal that tafgor influences the growth and IAA and siderophore production by soil bacteria, which could affect the plant growth and also bacterial diversity.

## CONCLUSION

The application of tafgor pesticide in the tomato crop field soil could elicit deleterious effect on soil microbial activity. This study focuses on the impact of tafgor at 15 ppm and 30 ppm on the growth of *Bacillus licheniformis*, *Bacillus cereus* and *Paenibacillus polymyca* and plant growth promoting traits of these bacteria like IAA and siderophore production. The results obtained revealed that tafgor significantly decreased the growth of bacteria in a dose-dependent manner. Consequently, significantly reduced the production of IAA by these bacteria. Thus, the persistence of tafgor and its residues could reduce the bacterial biodiversity and also effect the plant growth.

## LITERATURE CITED

1. Javaid MK, Ashiq M, Tahir M. 2016. Potential of biological agents in decontamination of agricultural soil. *Scientifica* 1-9.
2. Adhikari D, Perwira IY, Araki KS, Kubo M. 2016. Stimulation of soil micro-organisms in pesticide-contaminated soil using organic materials. *AIMS Bioengineering* 3(3): 379-388.
3. Susilowati DN, Riyanti EI, Setyowati M, Mulya K. 2002. Indole-3-Acetic acid producing bacteria and its application on the growth of rice. *AIP Conf. Proc* 020016-1-020016 -9.
4. Ahmad F, Ahamad I, Khan MS. 2008. Screening of free-living rhizospheric bacteria for their multiple plant growth promoting activities. *Microbiological Research* 163: 173-181.
5. Etesami H, Alikhani HA, Akbari AA. 2009. Evaluation of plant growth hormones production (IAA) ability by Iranian soil rhizobial strains and effects of superior strains application on wheat growth indexes. *World Applied Sciences Journal* 6(11): 1576-1584.
6. Fallik E, Okon Y. 1989. Identification and quantification of IAA and IBA in *Azospirillum brasilense* inoculated maize roots. *Soil Biol. Biochemistry* 21(1): 147-153.
7. Lwin KM, Myint MM, Tar T, Aung WZM. 2012. Isolation of plant hormone (indole-3-acetic acid- IAA) producing rhizobacteria and study on their effects on maize seedling. *Engineering Journal* 16(5): 137-144.

8. Park JM, Radhakrishnan R, Kang SM, Lee IJ. 2015. IAA producing *Enterobacter* sp. I-3 as a potent bio-herbicide candidate for weed control: a special reference with lettuce growth inhibition. *Indian Jr. Microbiology* 55(2): 207-212.
9. Khan AL, Halo BA, Elyassi A, Ali S, Hosni KA, Javid H, Harrasi AA, Lee IJ. 2016. Indole acetic acid and ACC deaminase from endophytic bacteria improves the growth of *Solanum lycopersicum*. *Electronic Journal of Biotechnology* 21: 58-64.
10. Raaska L, Viikari L, Sandholm MT. 1993. Detection of siderophores in growing cultures of *Pseudomonas* spp. *Journal of Industrial Microbiology* 11: 181-186.
11. Soni R, Kapoor R, Kaur M. 2016. Evaluation of siderophore production and antimicrobial activity by fluorescent *Pseudomonas* diversity associated with rhizosphere of apple and pear. *International Journal of Agriculture, Environment and Biotechnology* 9(6): 1109-1115.
12. Yeole RD, Dave BP, Dube HC. 2001. Siderophore production by fluorescent *Pseudomonas* colonizing roots of certain crop plants. *Indian Journal of Experimental Biology* 39: 464-468.
13. Gordon S, Weber RP. 1951. Colorimetric estimation of indole acetic acid. *Plant Physiology* 26: 190-95.
14. Brick JM, Bostock RM, Silverstone SE. 1991. Rapid in situ assay for indole acetic acid production by bacteria immobilized on a nitrocellulose membrane. *Applied and Environmental Microbiology* 57(2): 535-538.
15. Meyer JM, Abdallah MA. 1978. The fluorescent pigment of *Pseudomonas fluorescens*: biosynthesis, purification and physicochemical properties. *Journal of General Microbiology* 107: 319-328.
16. Shetti AA, Kaliwal BB. 2012. Imidacloprid induced intoxication in soil isolate *Brevundimonas* sp. MJ15. *International Journal of Life Science and Pharma Research* 2(3).
17. Shetti AA, Kulkarni AG, Kaliwal RB, Shivasharana CT, Kaliwal BB. 2012. Influence of imidacloprid on biochemical parameters in soil isolate *Escherichia coli*. *Int. Jr. Pharm. Bio. Sci.* 3(4): 1155-1163.
18. Tisler T, Jemec A, Mozetic B, Trebse P. 2009. Hazard identification of imidacloprid to aquatic environment. *Chemosphere* 76: 907-914.
19. Shetti A, Kaliwal BB. 2017. Effect of imidacloprid intoxication on growth and phosphatase activity in soil isolate *Bacillus weihenstephanensis* strain. *IOSR Journal of Biotechnology and Biochemistry* 3(4): 53-56.
20. Ahemad M, Khan MS. 2012. Effects of pesticides on plant growth promoting traits of *Mesorhizobium* strain MRC4. *Journal of the Saudi Society of Agricultural Sciences* 11: 63-71.
21. Mohite B. 2013. Isolation and characterization of indole acetic acid (IAA) producing bacteria from rhizospheric soil and its effect on plant growth. *Journal of Soil Science and Plant Nutrition* 13(3): 638-649.
22. Ali B, Sabri AN, Ljung K, Hasnain S. 2009. Auxin production by plant associated bacteria: impact on endogenous IAA content and growth of *Triticum aestivum* L. *Letters in Applied Microbiology* 48: 542-547.
23. Patten CL, Glick BR. 1996. Bacterial biosynthesis of indole-3-acetic acid. *Canadian Journal of Microbiology* 42: 207-220.
24. Wagi S, Ahmed A. 2019. *Bacillus* spp.: potent microfactories of bacterial IAA. *Peer Journal* 7: e7258
25. Ghosh SK, Pal S, Chakraborty N. 2015. The qualitative and quantitative assay of siderophore production by some microorganisms and effect of different media on its production. *Int. Jr. Chem. Sci.* 13(4): 1621-1629.
26. Kannahi M, Senbagam N. 2014. Studies on siderophore production by microbial isolates obtained from rhizosphere soil and its antibacterial activity. *Journal of Chemical and Pharmaceutical Research* 6(4): 1142-1145.
27. Kumar V, Sing S, Upadhyay N. 2019. Effects of organophosphate pesticides on siderophore producing soil microorganisms. *Biocatalysis and Agricultural Biotechnology* 21: 101359.
28. Ahemad M, Khan MS. 2011. Assessment of plant growth promoting activities of rhizobacterium *Pseudomonas putida* under insecticide-stress. *Microbiology Journal* 1(2): 54-64.
29. Chennappa G, Purushothama ACR, Naik MK, Suraj U, Sreenivasa MY. 2014. Impact of pesticides on PGPR activity of *Azotobacter* sp. isolated from pesticide flooded paddy soils. *Greener Journal of Agricultural Sciences* 4(4): 117-129.
30. Aswathi S, Gade RM, Shitole AV, Kapali S, Yogeshwar M. 2016. Studies on tolerance and sensitivity of fungal and bacterial bioagents to three pesticides commonly used in agriculture. *An International Quarterly Journal of Environmental Sciences* 9: 775-781.
31. Panomkhum P, Rungthong R, Nittayachit T, Iwai CB, Oun MT, Lawongsa P. 2014. Effects of pesticide on phenotypic characteristics of plant growth-promoting rhizobacteria (PGPR) in cassava production systems. *International Journal of Environmental and Rural Development* 5(1): 125-130.
32. Tripti, Kumar A, Kumar V, Anshumali. 2015. Effect of commercial pesticides on plant growth promoting activities of *Burkholderia* sp. strain L2 isolated from rhizosphere of lycopersicon esculentum cultivated in agricultural soil. *Toxicological and Environmental Chemistry* 97(9): 1180-1189.
33. Swarupa P, Kumar A. 2018. Impact of chlorpyrifos on plant growth promoting rhizobacteria isolated from *Abelmoschus esculentus*. *Jr. Pure Appl. Microbiology* 12(4): 2149-2157.
34. Kumar VS, Menon S, Agarwal H, Divya G. 2017. Characterization and optimization of bacterium isolated from soil samples for the production of siderophores. *Resource-Efficient Technologies* 3: 434-439.