



RESEARCH ARTICLE

INFLUENCE OF MICROBIAL INOCULANTS ON TRICHOMES IN TOMATO AGAINST FRUIT WORM

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ABSTRACT

The influence of microbial inoculants on the entrapment and impedance of fruit worm, *Helicoverpa armigera* (Hubner) in an already identified insect tolerant, tomato accession Varushanadu Local in comparison with a susceptible check, I 979 was studied under glasshouse conditions at Department of Entomology, Faculty of Agriculture, Annamalai University. Among the microbial inoculants applied plants, K solubilizer treated plants of Varushanadu Local entrapped the neonates caused the maximum mortality. In impedance tests *H. armigera* larvae took the maximum time on the foliage of plants supplied with K solubilizer followed by phosphobacteria.

Key words: Tomato, Entrapment, Impedance, Microbial inoculants, *H. armigera*.

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INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill) is the most widely grown solanaceous vegetable crop. Among the various insect pests responsible for lowering the yield of tomato, the fruitworm, *Helicoverpa armigera* (Hubner) is a highly destructive pest causing serious damage (Krishnamoorthy and Mani, 1996). Among the bio-physical factors of tomato, trichome density on the foliage was found to exert a profound influence on the insect activity. Trichomes are a common anatomical feature of the leaves and petioles of many crop plants including tomato which were reported to offer resistance against certain insect pests (Selvanarayanan and Narayanasamy, 2006). Keeping this point in view, the present investigation was carried out to analyse the role of trichomes in interrupt the neonates of *H. armigera* on selected tomato accessions as influenced by microbial inoculants.

MATERIAL AND METHODS

Based on preliminary and confirmatory field screening of 321 tomato accessions for resistance against fruit worm *H. armigera*, a promising accession Varushanadu Local was selected (Selvanarayanan and Narayanasamy, 2004) for further studies on the influence of microbial inoculants in enhancing resistance traits. For comparison, a susceptible check, I 979 was also evaluated. The evaluation was conducted under Glasshouse condition at the Department of Entomology,

Faculty of Agriculture, Annamalai University. The mean average temperature and relative humidity during these seasons were 28°C to 33°C and 70% to 85% respectively. For raising the seedlings, earthen pots of 30cm diameter were filled with potting mixture comprising two parts of soil, one part of sand and one part of farm yard manure. Then the seeds were sown and covered with a thin layer of sand. The seedlings were irrigated regularly. Twenty five days old seedlings were transplanted @ one seedling per pot. For induction of resistance in tomato accessions by microbial inoculations namely, *Azospirillum*, *Pseudomonas*, Phosphobacteria obtained from the Department of Agricultural Microbiology, Annamalai University and K- solubilizer (*Frateuria aurentia*) obtained from Romvijay Biotech Limited, Puducherry, India were used.

Estimation of density and types of trichomes

Density and types of trichomes present in the adaxial surfaces of the leaf was estimated. One mm long transverse section was cut from the leaf of the accessions. Then the sectioned sample was placed transversely on a clean glass slide and the number of trichomes were counted using a binocular microscope and expressed as trichome density per one mm length. (Kauffman and Kennedy, 1989)

Entrapment experiment

Young, fully expanded leaflets from 35 days old test plants were excised and placed individually, adaxial side up on a moist filter paper spread at the bottom of 80 mm plastic petridish. On each leaflet, 10 neonates were placed using a fine camel hair brush, on the adaxial leaf surface and the lid was

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S. No.	Treatments	Dosage / Pot	Day of application	Method of application
1.	<i>Azospirillum</i>	200 mg	On the day of transplanting	Soil
2.	Phosphobacteria	200 mg	On the day of transplanting	Soil
3.	<i>Pseudomonas</i>	200 mg	On the day of transplanting	Soil
4.	K – solubilizer (<i>F. aurentia</i>)	3 ml/kg of seed	One day before sowing	Seed treatment

placed on top to avoid desiccation. The larvae were gently prodded with a camel hair brush at 12 hrs after placement. If no reaction was evident, the neonate was designated trapped and dead. As control, leaflets excised from each test plant were gently swabbed on both sides using cotton moistened with 95 per cent ethanol to break the trichome heads and to remove the trichome exudates. These leaflets were then rinsed in distilled water to remove the ethanol. Five replications were maintained and ten neonates were used per replication (Simmons *et al.*, 2004).

Impedance experiment

Fully expanded leaflets from 35 days old test plants were excised and placed individually adaxial side up on a foam sheet. Two foam strips were kept on the foam sheet parallel to each other leaving a gap of one cm. The inner sides of the foam strips were smeared with wax to avoid larval climbing. One third instar larva was allowed to crawl on the leaf between the foam strips from one end to another and the time taken by the larva was recorded. Five replications were maintained at the rate of ten larvae per replication. As control, leaflets excised from each test plant were gently swabbed on both sides using cotton moistened with 95 per cent ethanol to break the trichome heads and to remove the trichome exudates. These leaflets were then rinsed in distilled water to remove the ethanol.

Statistical analysis

All the experiments were conducted in a completely randomized design and analysis of variance was used to work out the critical difference by adopting the procedure stated by Gomez and Gomez (1984).

RESULTS

Results of the experiments conducted to study the interaction of *Helicoverpa armigera* (Hubner) neonates and adaxial leaf surface of tomato accessions as influenced by microbial inoculants are presented hereunder.

Density and types of trichomes on adaxial surface

In the adaxial foliage surface of the tomato accessions, glandular and non-glandular trichomes were observed. Four types of trichomes such as, type I, a tall elongated multicellular stalk, type IV, a short multicellular stalk with a monocellular base, type VI, a multicellular stalk with a 2-4 cellular glandular head and monocellular base and type VII, a very short unicellular stalk with a 4 - 8 celled glandular head were detected in the leaves of accessions. Data on density and types of trichomes present in the adaxial leaf surface of the accessions as influenced by microbial inoculants are presented in Table 1.

Table 1. Trichome density on the adaxial leaf surface of the tomato accessions as influenced by microbial inoculants

S. No.	Treatments	Type I		Type IV		Type VI		Type VII		Total	
		VL	I 979	VL	I 979	VL	I 979	VL	I 979	VL	I 979
1.	<i>Azospirillum</i>	19.20 ±2.64	18.10 ±2.93	9.00 ±1.84	9.53 ±1.94	2.07 ±0.83	1.97 ±1.10	0.80 ±0.66	1.50 ±0.63	33.00	27.80
2.	Phosphobacteria	16.17 ±3.04	14.57 ±4.07	7.07 ±1.62	9.03 ±2.24	2.43 ±1.45	1.77 ±0.94	1.73 ±0.91	1.53 ±0.92	34.14	29.14
3.	<i>Pseudomonas</i>	13.97 ±2.44	13.37 ±3.55	10.40 ±2.11	9.47 ±2.36	1.10 ±0.66	2.47 ±1.11	1.40 ±0.89	1.23 ±0.86	28.20	26.54
4.	K-Solubilizer	19.73 ±2.50	18.63 ±3.25	9.47 ±1.94	9.67 ±1.71	2.43 ±1.07	2.27 ±1.23	1.57 ±0.90	1.30 ±1.04	36.80	31.10
5.	Control	15.20 ±2.26	13.00 ±3.90	7.37 ±2.47	7.73 ±2.07	2.23 ±1.10	1.73 ±1.11	1.07 ±1.09	1.23 ±0.57	21.86	23.69

Each value is a mean of thirty replications
Mean values followed by standard deviation

Table 2. Entrapment of *H. armiger* neonates on the tomato accessions as influenced by microbial inoculants

S. No.	Treatments	Larval mortality (%) after 12 hrs			
		VL		I 979	
		Trichome present	Trichome removed	Trichome present	Trichome removed
1.	<i>Azospirillum</i>	6.6 (23.74)	6.6 (12.59)	13.3 (21.14)	10.0 (18.43)
2.	Phosphobacteria	36.6 (36.93)	20.0 (26.56)	33.3 (35.21)	3.3 (6.74)
3.	<i>Pseudomonas</i>	16.6 (23.36)	6.6 (12.59)	13.3 (21.14)	13.3 (21.14)
4.	K-Solubilizer	43.3 (41.07)	20.0 (26.56)	26.6 (30.78)	16.6 (23.85)
5.	Control	6.6 (23.36)	10.0 (18.43)	10.0 (18.43)	10.0 (18.43)

CD (p=0.05) 7.49 7.147.826.92

Each value is a mean of three replications

Ten neonates used per replication

Table 3. Impedance of *H. armiger* larva on the tomato accessions as influenced by microbial inoculants

S. No.	Treatments	Time taken by larva on the accession (Sec)			
		VL		I 979	
		Trichome present	Trichome removed	Trichome present	Trichome removed
1.	<i>Azospirillum</i>	12.30 ±2.36	9.40 ±2.67	10.10 ±3.28	6.80 ±1.99
2.	Phosphobacteria	11.00 ±5.42	10.50 ±3.41	14.90 ±4.23	8.20 ±2.16
3.	<i>Pseudomonas</i>	10.30 ±1.25	6.20 ±2.04	9.00 ±2.62	6.80 ±1.55
4.	K-Solubilizer	15.40 ±3.34	6.80 ±1.55	12.40 ±1.96	8.00 ±1.89
5.	Control	10.00 ±1.25	8.10 ±2.02	8.00 ±5.42	6.40 ±1.42

Each value is a mean of ten replications

Mean values followed by standard deviation

The accession Varushanadu Local had the maximum number of all type trichomes irrespective of the treatments. Among the treatments, K solubilizer treated plants had the maximum number of trichomes in both accessions. This was followed by plants nourished with Phosphobacteria in case of both the accessions. Among the types of trichomes, number of Type I and IV trichomes was the maximum in the plants treated with K solubilizer. Type VI and type VII trichomes were predominant in phosphobacteria treated plants of the both accessions.

Entrapment of *H. armigeraneonates*

To analyse the influence of trichome type and density on *H. armigeraneonates*, entrapment test was conducted. When the neonates were allowed to move on the leaf surface, after 12 hrs of release, mortality rate was the maximum in the plants of Varushanadu Local treated with K solubilizer. But in I 979 plants, the maximum mortality occurred on the plants nourished with phosphobacteria. Whereas in case of trichomes removed leaf surface, the maximum mortality occurred on the foliage of Ksolubilizer and phosphobacterianourished plants of Varushanadu Local. In case of I 979, the maximum mortality occurred on trichome removed foliage of phosphobacteria treated plants (Table 2).

Impedance of *H. armigeralarvae*

On estimating the influence of trichome type and density of the accessions as influenced by microbial inoculants on movement of third instar of *H. armigeralarvae*, it was observed that the larvae took the maximum time on the foliage of plants supplied with K solubilizer. The larvae took the maximum time in case of the plants of the accession I 979 supplied with phosphobacteria. On trichomes removed leaf surface, larvae took the maximum time on the foliage of K solubilizer applied plants of Varushanadu Local. In case of I 979, the trichome removed foliage of phosphobacteria nourished plants impeded the movement of larvae to the maximum (Table 3).

DISCUSSION

To analyse the influence of trichome types and density on *H. armigeraneonates*, entrapment test was conducted. The maximum mortality of *H. armigeraw* was recorded in case of K solubilizer treated plants. Similarly, Juvik *et al.* (1994) evidently proved that the presence of high level of toxic acyl sugars in glandular trichomes exudates play a major role in the resistance of *Lycopersicon pennellii* to tomato fruit worm *Helicoverpa zea* Boddie. The K solubilizer induced resistance resulted in enhanced level of phenol production in the tomato plants. Phenol compound has been implicated as a possible factor in inhibiting growth and development of *H. zea* larvae (Isman and Duffey, 1982a). The phenol and chlorogenic acid in the leaf lamella and tips of glandular trichomes account for over 60 per cent of the total phenol content of tomato (Isman and Duffey, 1982b). Trichome density on the adaxial surface was found to have a significant negative correlation with the larval mortality on the foliage of both accessions. On estimating the influence of trichomes on the movement of *H. armigeralarvae* on foliage surface, it was observed that the larvae took the maximum time on the foliage of plants supplied with K solubilizer. This is may due to maximum number of non

glandular (Type I) and glandular (Type VII) trichomes present on the foliage of plants nourished with K solubilizer. Trichome density on the adaxial surface was found to have a significant negative correlation with the larval movement on the foliage of both accessions. Simmons *et al.* (2003) reported that non-glandular trichomes affect pests by providing mechanical barrier to movement or access to nutritious tissues. Simmons *et al.* (2004) stated that glandular trichomes also arrest the movement of herbivores by means of the release a sticky and/or toxic exudates that has the potential to trap an arthropod on contact, leading to its death via starvation or mortality as a result of toxins.

Conclusion

It is concluded from the present investigation that the accession Varushanadu Local was less preferred by *H. armigera*. The maximum mortality of *H. armigeraw* was recorded in case of K solubilizer treated plants and larvae took the maximum time on the foliage of plants supplied K-solubilizer.

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