Induction of plant immune by microbial inoculants against tomato fruit worm

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ABSTRACT

Tomato (*Lycopersicon esculentum* Mill.) is one of the important and remunerative vegetable crops grown around the world for fresh market and processing. The production and productivity of the crop is greatly hampered by the fruit borer, *Helicoverpa armigera* (Hübner) which causes damage to the developing fruits and results in yield loss. The indiscriminate use of synthetic chemical pesticides to control this pest resulted in development of resistance and harmful pesticide residues in fruits. To avoid such problems caused due to indiscriminate use of insecticides, utilization of Host Plant Resistance (HPR) is an ecologically viable, alternate insect pest management strategy. The tomato accession Varushanadu Local along with a popular variety PKM1 was tested for induction of resistance by various microbial inoculants *viz., Azospirillum,* Phosphobacteria, *Pseudomonas,* K-solubilizer, *Azospirillum* + Phosphobacteria, *Azospirillum* + *Pseudomonas, Azospirillum* + K-solubilizer, Phosphobacteria + *Pseudomonas,* Phosphobacteria + K - solubilizer. and *Pseudomonas.* Based on the preliminary study, the promising microbial inoculants *viz.*, *Pseudomonas*, K-solubilizer, *Azospirillum,* + k-solubilizer, *pseudomonas* + k-solubilizer, and phosphobacteria + K-solubilizer were selected. Feeding preference of *H. armigera* to the leaves and fruits was the minimum towards the accessions Varushanadu Local treated with *Pseudomonas* + K-solubilizer. Plants of the accessions Varushanadu Local supplied with *Pseudomonas* + K-solubilizer exerted profound antibiosis effects against the life stages of *H. armigera* as evidenced by higher larval mortality, reduced pupation, reduced larval, pupal and adult duration.

Keywords: Tomato, Biofertilizers, Feeding preference, Antibiosis

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INTRODUCTION

Tomato (*Lycopersicon esculentum*) an important vegetable crop in India. The most important insect pest of tomato is fruitworm, *Helicoverpa armigera* (L) which causes damage to the developing fruits and results in yield loss (Muthukumaran and Selvanarayanan, 2016). *H. armigera* is active throughout the year and its severe damage is noticed during fruiting periods of the crop. The newly hatched larvae feed on the foliage while older larvae bore the fruit for feeding (Khanam *et al*., 2003). The indiscriminate use of synthetic chemical pesticides to control this pest resulted in development of resistance and harmful pesticide residues in fruits. To avoid such problems caused due to indiscriminate use of insecticides, utilization of Host Plant Resistance (HPR) is an ecologically viable, alternate insect pest management strategy. In the absence of natural resistance in the gene pool of crop plants or lack of desirable yield attributes in the identified insect tolerant/ resistant crop varieties, inducing resistance by manipulation of plant nutrients may be attempted (Muthukumaran and Selvanarayanan, 2010). Keeping this in mind, the present study was undertaken to analyse the role of certain microbial inoculants in enhancing insect resistance in the selected tomato accessions.

MATERIALS AND METHODS

Based on preliminary and confirmatory field screening of 321 tomato accessions for resistance against fruitworm *H. armigera*, a promising accession Varushanadu Local was selected (Selvanarayanan and Narayanasamy, 2006) for further studies on the influence of biofertilizers in enhancing resistance traits. For comparison, a popular variety, PKM 1 was also evaluated. The evaluation was conducted under glasshouse condition at the Department of Entomology, Faculty of Agriculture, Annamalai University from July 2009 to October 2010. The mean average temperature and relative humidity during these seasons were 28^{n}C to 33°C and 70% to 85% respectively. For raising the seedlings, earthen pots of 30 cm 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-day old seedlings were transplanted @ one seedling per pot.

The influence of various microbial inoculants on the mechanisms of resistance namely preference non preference of *H. armigera* larvae for feeding on foliage and fruits and antibiosis of the tomato plants on the insect development were investigated. For induction of resistance in tomato accessions by microbial inoculation, the microbial inoculants 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. The details on various combinations of microbial inoculants evaluated in the present study are described hereunder.

Treatments	Dosage / Pot	Day of application	Method of application
Azospirillum(T1)	200 mg	On the day of transplanting	Soil
Phosphobacteria (T2)	200 mg	On the day of transplanting	Soil
Pseudomonas(T3)	200 mg	On the day of transplanting	Soil
K – solubilizer (*F. aurentia*) (T4)	3 ml/kg of seed	One day before sowing	Seed treatment
Azospirillum + Phosphobacteria (T5)	200mg+200mg	On the day of transplanting	Soil
Azospirillum + *Pseudomonas*(T6)	200mg+200mg	On the day of transplanting	Soil
Azospirillum + K – solubilizer (*F. aurentia*) (T7)	200 mg +3 ml/kg of seed	On the day of transplanting and one day before sowing	Soil + Seed treatment
Phosphobacteria + *Pseudomonas*(T8)	200mg+200mg	On the day of transplanting	Soil
Pseudomonas + K – solubilizer (*F. aurentia*)(T9)	200 mg +3 ml/kg of seed	On the day of transplanting and one day before sowing	Soil + Seed treatment
Phosphobacteria + K – solubilizer (*F. aurentia*)(T10)	200 mg +3 ml/kg of seed	On the day of transplanting and one day before sowing	Soil + Seed treatment
Control	–	–	–

Relative leaf and fruit damages by confined feeding

Single third instar larva of *Helicoverpa armigera* was pre starved for six hours and then allowed into a specially designed screening cage 10 days after transplanting to feed individually on the leaves of tomato plants treated with various microbial inoculants. The screening cage consisted of cylindrical major film sheet rolled tubular (10.5 cm dia and 25 cm long), open and were affixed with muslin cloth and nylon mesh cloth at each end. The cage was fixed on top of a wooden stick (70 cm high) three such replications were maintained per treatment and the area of the leaf infested by the larvae after 24,48 and 72 hours was measured. On the seventh day from the first fruit appearance, young fruits of the accessions treated with different microbial inoculants were excised with calyx. They were placed individually inside a plastic container with moist filter paper spread at the bottom. Fruit afresh. Three replications per treatment were maintained. Third instar larvae @ one per replications, pre-starved for six hours were allowed to feed on the fruits. After 24, 48 and hours, the reduction in initial fruit weight was recorded. A control set for each treatment was recorded. A control set for each treatment was also maintained without larval release to study the reduction in fruit weight due to drying of fruit consumed in the treatments with larval release.

Relative leaf and fruit damages by free choice

Relative preference of *H. armigera* larvae to leaves of the test plants was ascertained by leaf disc method (Kauffman and Kennedy, 1989). Leaf disc of 25mm^2 size was excised from second leaf beneath the terminal bud of 40 days old plants (10DAT) from each treatment and were placed at equal distant circularly on moist filter paper in a 150mm dia Petridis. Third instar larvae @ one per replication, pre starved for hours were allowed to feed. The leaf area consumed by the larvae after 24, 48 and 72 hours was measured using graph sheet. This experiment was replicated three times. Young fruits of the accessions treated with different microbial inoculants were excised with calyx weighed individually and placed at equal distant circularly in a plastic container (30 cm×15 cm×8 cm) having moist filter paper spread at the bottom @ one fruit per treatment. Three replications were maintained and 6hours pre-starved third instar larvae @ one per replications was released at the centre of the container. Reduction in the fruit weight after 24, 48 and 72 hours was recorded. A control set for each treatment was also maintained without larval release to study the reduction in fruit weight due to drying. This reduction in was taken into consideration while computing quantum of fruit consumed in the treatments with larval release.

Antibiosis of tomato accessions against *H.armigera*

The tomato accessions, as influenced by various microbial inoculants were tested for their antibiosis effects, if any, on *H.armigera.* Ten neonate larvae were released individually in the screening cage enclosing the foliage of the plants in each treatment for both the accessions, each treatment for both the accessions, each in five replications. Supplied with fresh foliage and fruit whenever needed and larval mortality, pupation rate, larval, pupal and adult longevity and adult emergence were recorded.

Statistical analysis

The data thus obtained from screening of tomato accessions treated with selected microbial inoculants were analyzed statistically using factorial randomized block design (FRBD). The data thus gathered were statistically analyzed using IRRISTAT 4.0 software and the critical difference values were arrived at.

RESULTS AND DISCUSSION

The foliage preference of *H. armigera* larvae, as influenced by microbial inoculants was observed maximum in the confinement test than free choice test. The feeding damage was more in PKM1 than Varushanadu Local, irrespective of treatments. In line with this, larval populations of the fruit worm *H. armigera* was found to be the least in Varushanadu Local as earlier reported by

Dhakshinamoorthy (2002) and Selvanarayanan and Narayanasamy (2006). The accession Varushanadu Local collected from a hilly terrain in Southern India is a suspected natural cross between *L. esculentum* and *L. pimpinellifolium* and hence the resistance traits derived from the wild accession *L. pimpinellifolium* would have offered such resistance. Such wild relatives or their derivatives have been reported to possess resistance against the fruit borer, *H. armigera* (Sankhyan and Verma, 1997).

In both confinement and free choice tests, among the treatments, *Pseudomonas* + K-solubilizer (T9) nourished accessions were less preferred by *H. armigera* larvae followed by K-solubilizer (T4) applied plants. *Pseudomonas* + K-solubilizer (T9) nourished PKM 1 recorded higher leaf damage whereas Varushanadu local was less preferred by *H. armigera* in confinement and free choice test respectively. In the all experiments, the accession PKM 1 was highly preferred by *H. armigera*. In glasshouse evaluation, accession Varushanadu Local was less preferred by *H. armigera*. As for fruit feeding preference, In confinement test, among this treatment, fruits of plants applied with combination of *Pseudomonas* + K-solubilizer were less preferred by *H. armigera* larvae in case of the accession Varushanadu local. Similarly, accession PKM1 supplied with various microbial inoculants, Plants supplied with combination of *Pseudomonas* + K-solubilizer (T9) recorded lesser fruit feeding preference by *H. armigera larva*. This was followed by fruits of plants applied with K-solubilizer alone (T4) . In free choice test also, among the treatments, in both the accessions, foliage of plant nourished with *Pseudomonas* + K-solubilizer (T9) combination were less preferred by *H. armigera* larvae. This was followed by foliage of plants treat with K-solubilizer only (Table 1).

With regard to the antibiosis effect of the tomato accessions as influenced by microbial inoculants on *H. armigera,* it was observed that, the accessions, Varushanadu Local exerted pronounced antibiosis effect on the life stages of *H. armigera* irrespective of the microbial inoculants. Among this microbial inoculants, combination of *Pseudomonas* + K-solubilizer nourished plants exerted higher antibiosis effect on *H.armigera*. *Pseudomonas* + K-solubilizer applied plants caused the maximum larval mortality. Regarding adult emergence, *Pseudomonas* + K-solubilizer (T9) nourished plants of the both accessions recorded the least adult emergence and pupation rate (Table 2). Similar negative influence of potassium on certain insect pests was reported by Marwat *et al*. (1985) who found the population of cabbage aphid to have negative correlation with increasing level of potassium. On the other hand, Inayatullah (1987) concluded that potassium had positive correlation with the sugarcane borer infestation. The increase in the population of K-solubilizing microbe in the rhizosphere would have enhanced the ready availability of potassium to the plants and in turn its higher uptake. This enhanced potassium content in the foliage would have evinced a negative influence on *H. armigera* as reported earlier by Marwat *et al*. (1985).

Table 1. Fruit feeding preference of *H. armigera* larvae towards tomato accessions as influenced by bioinoculants –Confinement & Free choice test

Treatments	Quantum of fruit (g) consumed at 72 hrs			
	Confinement		Free choice	
	VL	PKM 1	VL	PKM 1
T1	0.58	0.68	0.80	0.92
T2	0.56	0.76	0.74	0.84
T3	0.50	0.60	0.56	0.65
T4	0.44	0.54	0.40	0.51
T5	0.54	0.64	0.72	0.83
T6	0.52	0.62	0.70	0.80
T7	0.44	0.55	0.50	0.61
T8	0.50	0.60	0.68	0.79
T9	0.38	0.48	0.33	0.44
T10	0.42	0.52	0.44	0.54
T11	2.75	3.00	2.00	2.25

It is concluded from the present investigation that the accession Varushanadu Local was less preferred by *H. armigera*. Also, this accession evinced a higher antibiosis against *H. armigera*. Among the treatments, *Pseudomonas* + K-solubilizer (T9) nourished plants induced higher antibiosis effect on *H. armigera* as well as recorded lesser preference.

Table 2. Antibiosis effects of the tomato accessions on *H.armigera* as influenced by bioinoculants

Treatments	Larval mortality (%)		Pupation (%)		Adult emergence (%)	
	VL	PKM 1	VL	**PKM 1**	**VL**	**PKM 1**
T1	40.00	38.0	60.15	65.25	75.12	80.16
T2	42.00	40.00	65.18	70.45	78.36	82.17
T3	50.00	48.00	42.52	44.25	30.33	38.00
T4	56.00	52.00	45.12	40.12	30.18	35.52
T5	44.00	58.00	65.23	70.00	78.00	81.21
T6	48.00	46.00	58.00	60.02	55.00	45.02
T7	52.00	50.00	50.10	52.10	40.25	45.00
T8	46.00	40.00	56.00	60.00	52.23	44.23
T9	60.00	56.00	42.18	39.45	20.15	30.14
T10	54.00	51.00	40.00	38.02	25.00	30.00
T11	30.00	20.00	92.00	88.00	85.00	83.00

References

Dhakshinanoorthy, G. 2002. Studies on resistance of tomato against fruit borer, *Helicoverpa armigera* (Hubner). *M.Sc., (Ag.) Thesis***.** Annamalai University. India.

Inayatullah, C. 1987. Management of insects and weed pest in sugarcane and paddy crops in Pakistan. *Final technical report 1981-1987*. PL-480 project. NARC, Pak. Agric. Res. Council, Islamabad, 37 **P**.

Kauffman, W. C. and Kennedy, C. 1989. Relationship between trichome density in tomato and parasitism of *Heliothis* spp. eggs by *Trichogramma* spp., *Environmental Entomology***, 18:**698-704.

Khanam, U.K.S., Hussain, M. Ahmed, N. Uddin, M.M. and Hussain, M.S. 2003. Varietal screening of tomato to tomato fruit borer, *Helicoverpa armigera* (Hubner). and associated tomato plant characters. *Pakistan Journal of Biological Science* **6**(4): 413-421.

Marwat, G.A., Ahmed, I. and Khatoon, S. 1985. Role of different levels of potassium and population densities of cabbage aphids on sarson and its yield. *Journal of Agriculture*, **1(2):**85-89.

Muthukumaran, N. and Selvanarayanan, V. 2010. Differential reaction of hybrid derivatives of *Lycopersicon* accessions against whitefly *Bemisia tabaci* Genn. In South India. 235-238 **PP**.

Ignacimuthu, S.J. and David, B.V. (Eds.) Non-chemical Insect Pest Management, Elite Publishing House Pvt. Ltd., New Delhi, India. 266 **P**.

Muthukumaran, N. and Selvanarayanan, V. 2016. Influence of nutrient supply on antixenosis resistance in tomato accessions against *Helicoverpa armigera* (Hubner), *Annamalai University Science Journal*, **50**:123-126.

Sankhyan, S. and Verma, A.K. 1997. Field screening for resistance against the fruit borer *Helicoverpa armigera* (Hubner) (Lepidoptera:Noctuidae). *Pest Mangement and Economic Zoology*, **5 (2):** 107-111.

Selvanarayanan, V. and Narayanasamy, P. 2006. Factors of resistance in tomato accessions against the fruit worm *Helicoverpa armigera* (Hubner). *Crop Protection*, **256 (25):** 1075-1079.

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