



## Evaluation of Entomopathogenic Fungi against Two Spotted Spider mite, *Tetranychus urticae* Koch on Tomato

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Entomopathogenic fungi belonging to six genera were evaluated for effectiveness against *Tetranychus urticae* Koch along with the standard check fenazaquin 10 EC at 1.5 ml<sup>-1</sup> in both pot culture and field experiments. *Beauveria bassiana* was highly effective in reducing the mite population in pot culture and two field experiments with 54.57, 54.52, 51.87 per cent, respectively after two rounds of spraying. It was followed by *Hirsutella thompsonii*, *Metarhizium anisopliae* and *Paecilomyces fumosoroseus* in their order of effectiveness. More than 70 per cent reduction in mite population was recorded with the acaricide fenazaquin in all the experiments conducted.

**Key words:** Tomato, Two spotted spider mite, *Tetranychus urticae*, Entomopathogenic fungi

Two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae), is a cosmopolitan and polyphagous species with great economic importance for crops in greenhouses and in the field. Tomato (*Lycopersicon esculentum* Mill. Solanaceae) is an important vegetable crop in which India is the second largest producer next to China with an area of 8.65 lakh hectares, annual production of 16.826 million MT (11.5% of total production) and 19.5 MT of productivity (nhb.gov.in/area-pro/database-2011). The spider mite cause considerable leaf damage and yield decrease to tomato crop plants (Hussey and Scopes, 1985). Chemical control is the main method of combating spider mites. As a result of the excessive use of pesticides and the associated problems of pesticide resistance and environmental pollution, there is an increasing demand for sustainable, environmentally friendly control methods. Screening for eligible biocontrol agents is a step in developing new or improving existing environment-friendly strategies offering an alternative to conventional pest control. Acari make good hosts for fungal pathogens because they are generally soft bodied and many inhabit environments with humid microclimates (Evans, 1992) which favour infection and disease transmission (Hajek and St Leger, 1994). Keeping this view, the present study was undertaken to study the effectiveness of different fungal pathogens for the management of *T. urticae*.

### Materials and Methods

Six fungi of entomopathogenic Hyphomycetes were used in this study. Pure cultures of the entomopathogenic fungi, *Beauveria bassiana* (Balsamo) Vuillemin, from Department of

Agricultural Entomology, *Hirsutella thompsonii* (Fisher) and *Paecilomyces fumosoroseus* (Thom.) were obtained from the National Bureau of Agriculturally Important Insects (NBAll), Bangalore. *Metarhizium anisopliae* (Metchinkoff) Sorokin, *Cladosporium cladosporioides* (Fresenius) de Vries and *Fusarium pallidoroseum* Cooke (Sacc) from Department of Agricultural Entomology, TNAU, Coimbatore. Fungi were mass multiplied in coconut water in the laboratory for spray application. 300 ml of 50 per cent coconut water was poured in 500 ml round bottom flask and autoclaved at 121°C for 20 minutes. After cooling, one ml of spore suspension of fungal pathogens were inoculated into each flask separately and incubated at room temperature for 15 days. Spore count in each flask was estimated using a Neubauer haemocytometer. The standard check used was fenazaquin at 1.5ml<sup>-1</sup> and a control plot was maintained.

Pot culture experiment was conducted at Insectary glasshouse, TNAU, Coimbatore. Two field experiments were conducted in farmers' field one at Puttur (November 2011 to January 2012) and another at Thondamuthur (March to May, 2012) in Coimbatore district of Tamil Nadu. The experiments were carried out in a randomized block design with the plot size of 4m x 5m. All the eight treatments were replicated thrice. The fungal cultures were applied on plants in the evening hours using a hand sprayer for pot culture experiment and knapsack sprayer for field experiments. The pre and post treatment observations on live mite population were assessed on 0, 3, 7, 10 and 14 days after application from five plants selected randomly in each plot. Three leaves one each from top, middle and bottom canopies of each plant were assessed for the live mites (nymphs and adults) on the undersurface in

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an area of 4cm<sup>2</sup>. The observations were made randomly on the place where maximum population was noticed. The mite population was assessed using 10x hand lens. Treatments were imposed at 14 days interval.

The data were transformed and analysed statistically. The mite population from the pot culture and field experiments were subjected to square root (X+0.5) transformation. The analysis of variance in different experiments was carried out in AGRES (ver 7.01 Pascal International Software Solutions, USA) and the means were separated by least significant difference (LSD) available in the package.

## Results and Discussion

In pot culture experiment, the mite population before imposing treatments ranged from 28.22 to 31.36 mites per 4 cm<sup>2</sup> and was statistically non significant. After first spraying, *B. bassiana* at 10<sup>8</sup> spores ml<sup>-1</sup> was significantly superior which recorded the lowest mite population of 21.47, 20.85, 16.62 and 18.58 mites per 4 cm<sup>2</sup> at 3,7,10 and 14 days after spraying, respectively. It recorded the highest mean population reduction of 40.71 per cent, followed by *H. thompsonii* with 38.68 per cent (Table 1). After the second round of spraying, a similar trend was observed. Spraying of *B. bassiana*

**Table 1. Efficacy of entomopathogenic fungi against *T. urticae* on tomato after two rounds of spraying Pot culture experiment**

| Treatment<br>(x 10 <sup>8</sup> Spores ml <sup>-1</sup> ) | PTC   | I Spraying                    | II Spraying                  | Pooled Mean                  | Cumulative %<br>reduction over<br>control |
|-----------------------------------------------------------|-------|-------------------------------|------------------------------|------------------------------|-------------------------------------------|
|                                                           |       | Mean no. of mites             | Mean no. of mites            |                              |                                           |
| <i>B. bassiana</i>                                        | 29.19 | 19.38<br>(4.46) <sub>b</sub>  | 11.40<br>(3.45) <sub>b</sub> | 15.39<br>(3.99) <sub>b</sub> | 54.57                                     |
| <i>M. anisopliae</i>                                      | 30.20 | 23.49<br>(4.90) <sub>c</sub>  | 15.91<br>(4.05) <sub>d</sub> | 19.70<br>(4.49) <sub>b</sub> | 45.34                                     |
| <i>P. fumosoroseus</i>                                    | 27.93 | 22.24<br>(4.77) <sub>bc</sub> | 14.07<br>(3.82) <sub>e</sub> | 18.16<br>(4.32) <sub>b</sub> | 47.33                                     |
| <i>H. thompsonii</i>                                      | 28.37 | 20.05<br>(4.53) <sub>b</sub>  | 12.21<br>(3.57) <sub>c</sub> | 16.13<br>(4.08) <sub>b</sub> | 52.33                                     |
| <i>F. pallidroseum</i>                                    | 30.89 | 25.95<br>(5.14) <sub>c</sub>  | 17.51<br>(4.24) <sub>g</sub> | 21.73<br>(4.71) <sub>b</sub> | 39.13                                     |
| <i>C. cladosporioides</i>                                 | 31.36 | 23.49<br>(4.90) <sub>c</sub>  | 17.53<br>(4.25) <sub>f</sub> | 20.51<br>(4.58) <sub>b</sub> | 41.65                                     |
| Fenazaquin 10 EC 1.5 ml <sup>-1</sup>                     | 28.22 | 12.78<br>(3.64) <sub>a</sub>  | 5.03<br>(2.35) <sub>a</sub>  | 8.90<br>(3.07) <sub>a</sub>  | 73.60                                     |
| Control                                                   | 28.87 | 32.69<br>(5.76) <sub>d</sub>  | 37.74<br>(6.18) <sub>h</sub> | 35.22<br>(5.98) <sub>c</sub> | 0.00                                      |
| SEd                                                       |       | 0.1772                        | 0.1085                       | 0.371                        |                                           |
| CD(0.05)                                                  |       | 0.3684                        | 0.2256                       | 0.878                        |                                           |

In a column means followed by a common letter (s) are not significantly different at P = 0.05 by LSD Figures in parentheses are "x+0.5 transformed values. PTC- Pretreatment count

@ 10<sup>8</sup> spores ml<sup>-1</sup> recorded the highest cumulative mean population reduction of 54.57 per cent followed by *H. thompsonii* and *P. fumosoroseus* which recorded a mean reduction of 52.33 and 47.33 per cent, respectively. The standard check, fenazaquin 10 EC (1.5 ml lit<sup>-1</sup>) was significantly superior to all other treatments and recorded the highest cumulative population reduction of 73.60 per cent (Table 2). The results are in tune with the findings of Ghosh *et al.* (2007) who showed that *B. bassiana* (85.47%), *P. fumosoroseus* (84.22%) and *H. thompsonii* (80.47%) were effective against *T. urticae* on French beans. Chandler *et al.* (2005) recorded Naturalis-L (*B. bassiana*) reduced *T. urticae* populations up to 97 per cent. Afifi *et al.* (2010) showed 78.4 per cent reduction of *T. urticae* on cotton using *B. bassiana*. Gatarayiha *et al.*, (2012) recorded more than 80 per cent mortality in all the three tested strains of *B. bassiana* against *T. urticae*.

The result of the field experiment conducted in Puttur indicated that after two rounds of spraying *B.*

*bassiana* and *H. thompsonii* caused maximum cumulative mean per cent reduction of 54.52 per cent and 48.26 per cent, respectively (Table 3). Muthukumar (2005) recorded a population reduction of 55.22 per cent with *B. bassiana* against *T. urticae* on okra. The next effective treatments were *M. anisopliae* and *P. fumosoroseus* with a population reduction of 43.01 and 38.64 per cent, respectively. The fungal pathogens were effective up to 7 days, after which a decreased trend in the pathogenicity was recorded in two rounds of spraying. Fenazaquin 10 EC (1.5 ml lit<sup>-1</sup>) was significantly superior which recorded the highest population reduction of 75.15 per cent (Table 4). *C. cladosporioides* and *F. pallidroseum* were next in the order of efficacy with the cumulative population reduction of 34.20 and 30.60, respectively. The same trend was observed in the second field experiment in which after two rounds of spraying among the fungal pathogens, *B. bassiana* recorded the highest cumulative per cent reduction of 51.87 per cent followed by *H. thompsonii*

**Table 2. Efficacy of entomopathogenic fungi against *T. urticae* on tomato after two rounds of spraying Trial I (Puttur)**

| Treatment<br>(x 10 <sup>8</sup> Spores ml <sup>-1</sup> ) | PTC   | I Spraying                    | II Spraying                   | Pooled Mean                    | Cumulative %<br>reduction over<br>control |
|-----------------------------------------------------------|-------|-------------------------------|-------------------------------|--------------------------------|-------------------------------------------|
|                                                           |       | Mean no. of mites             | Mean no. of mites             |                                |                                           |
| <i>B. bassiana</i>                                        | 39.34 | 22.60<br>(4.81) <sub>b</sub>  | 13.87<br>(3.79) <sub>b</sub>  | 18.24<br>(4.33) <sub>b</sub>   | 54.52                                     |
| <i>M. anisopliae</i>                                      | 37.99 | 25.45<br>(5.09) <sub>c</sub>  | 20.41<br>(4.57) <sub>c</sub>  | 22.93<br>(4.84) <sub>bcd</sub> | 43.01                                     |
| <i>P. fumosoroseus</i>                                    | 40.14 | 27.65<br>(5.31) <sub>cd</sub> | 21.70<br>(4.71) <sub>bc</sub> | 24.68<br>(5.02) <sub>bcd</sub> | 38.64                                     |
| <i>H. thompsonii</i>                                      | 44.07 | 25.09<br>(5.06) <sub>bc</sub> | 16.43<br>(4.11) <sub>b</sub>  | 20.76<br>(4.61) <sub>bc</sub>  | 48.26                                     |
| <i>F. pallidoroseum</i>                                   | 38.02 | 29.61<br>(5.49) <sub>d</sub>  | 26.30<br>(5.18) <sub>c</sub>  | 27.96<br>(5.33) <sub>d</sub>   | 30.60                                     |
| <i>C. cladosporioides</i>                                 | 41.76 | 27.94<br>(5.33) <sub>cd</sub> | 25.08<br>(5.06) <sub>c</sub>  | 26.51<br>(5.20) <sub>cd</sub>  | 34.20                                     |
| Fenazaquin 1.5 ml <sup>-1</sup>                           | 36.46 | 13.33<br>(3.72) <sub>a</sub>  | 6.54<br>(2.65) <sub>a</sub>   | 9.94<br>(3.23) <sub>a</sub>    | 75.15                                     |
| Control                                                   | 36.96 | 39.29<br>(6.31) <sub>e</sub>  | 41.46<br>(6.48) <sub>e</sub>  | 40.38<br>(6.39) <sub>e</sub>   | 0.00                                      |
| SEd                                                       |       | 0.1333                        | 0.1956                        | 0.309                          |                                           |
| CD(0.05)                                                  |       | 0.2772                        | 0.4068                        | 0.730                          |                                           |

In a column means followed by a common letter (s) are not significantly different at P = 0.05 by LSD Figures in parentheses are "x+0.5 transformed values. PTC- Pretreatment count

with 45.06 per cent. *M. anisopliae* and *P. fumosoroseus* were on par in reducing the mite population to 34.30 and 28.41 per cent, respectively (Table 6). The statistical analysis on the interaction

effect revealed that the interaction between number of sprays, days after treatment and treatments were highly significant at one per cent level of probability. The treatment and spray interaction was highly

**Table 3. Efficacy of entomopathogenic fungi against *T. urticae* on tomato after two rounds of spraying Trial II (Thondamuthur)**

| Treatment<br>(x 10 <sup>8</sup> Spores ml <sup>-1</sup> ) | PTC   | I Spraying                    | II Spraying                  | Pooled Mean                   | Cumulative %<br>reduction over<br>control |
|-----------------------------------------------------------|-------|-------------------------------|------------------------------|-------------------------------|-------------------------------------------|
|                                                           |       | Mean no. of mites             | Mean no. of mites            |                               |                                           |
| <i>B. bassiana</i>                                        | 45.32 | 28.64<br>(5.40) <sub>b</sub>  | 17.49<br>(4.24) <sub>b</sub> | 23.06<br>(4.85) <sub>b</sub>  | 51.87                                     |
| <i>M. anisopliae</i>                                      | 41.42 | 36.86<br>(6.11) <sub>c</sub>  | 26.08<br>(5.16) <sub>d</sub> | 31.47<br>(5.65) <sub>cd</sub> | 34.30                                     |
| <i>P. fumosoroseus</i>                                    | 38.33 | 37.57<br>(6.17) <sub>c</sub>  | 31.00<br>(5.61) <sub>e</sub> | 34.28<br>(5.90) <sub>cd</sub> | 28.41                                     |
| <i>H. thompsonii</i>                                      | 44.07 | 31.72<br>(5.68) <sub>bc</sub> | 20.93<br>(4.63) <sub>c</sub> | 26.32<br>(5.18) <sub>c</sub>  | 45.06                                     |
| <i>F. pallidoroseum</i>                                   | 48.34 | 41.35<br>(6.47) <sub>c</sub>  | 37.95<br>(6.20) <sub>e</sub> | 39.65<br>(6.34) <sub>de</sub> | 17.17                                     |
| <i>C. cladosporioides</i>                                 | 46.30 | 38.01<br>(6.21) <sub>c</sub>  | 34.70<br>(5.93) <sub>e</sub> | 36.35<br>(6.07) <sub>d</sub>  | 24.06                                     |
| Fenazaquin 10 EC 1.5 ml <sup>-1</sup>                     | 42.50 | 14.36<br>(3.86) <sub>a</sub>  | 6.92<br>(2.72) <sub>a</sub>  | 10.64<br>(3.34) <sub>a</sub>  | 77.81                                     |
| Control                                                   | 41.94 | 48.10<br>(6.97) <sub>d</sub>  | 47.62<br>(6.94) <sub>f</sub> | 47.86<br>(6.95) <sub>e</sub>  | 0.00                                      |
| SEd                                                       |       | 0.2454                        | 0.810                        | 0.322                         |                                           |
| CD(0.05)                                                  |       | 0.5107                        | 0.3763                       | 0.762                         |                                           |

In a column means followed by a common letter (s) are not significantly different at P = 0.05 by LSD Figures in parentheses are "x+0.5 transformed values. PTC- Pretreatment count

significant on the fungal pathogens tested. Gatarayih (2010) reported that the control efficacy of *B. bassiana* on *T. urticae* was significantly different between crops such as beans, cucumber, egg plant, maize and tomato and their corresponding per cent

reduction were 56.7, 50.3, 51.7, 25.0 and 34.6, respectively that supports the variation in efficacy of *B. bassiana* among crops tested. Omar and Hanna (2002) reported *B. bassiana* recorded highest reduction in mite population to the tune (77.59%)

against *T. urticae* on cowpea. Vinoth kumar (2010) reported that the basal application of *Pseudomonas fluorescens* along with the two sprays of *B. bassiana* @10<sup>8</sup> spores ml<sup>-1</sup> recorded highest reduction of *T. urticae* population up to 71.99 per cent.

The results obtained in this study show that *B. bassiana* and *H. thompsonii* provided effective control of *T. urticae*. Considering that spider mite resistance to acaricides is a seriously increasing phenomenon, biopesticides may be a successful alternative to conventional chemical control. Repeated application of these entomofungal pathogens with suitable formulations for thriving under environmental conditions can be incorporated in the Integrated Pest Management (IPM) for the control of *T. urticae*.

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