



#### **Research Article**

# Biocontrol efficacy of entomopathogenic fungi against San Jose scale *Quadraspidiotus perniciosus* (Comstock) (Hemiptera: Diaspididae) in field trials

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**ABSTRACT:** San Jose scale *Quadraspidiotus perniciosus* is a key pest of apple crop in the northern states of India. The efficacy of entomopathogenic fungi - *Beauveria bassiana, Metarhizium anisopliae* sensu lato and *Lecanicillium lecanii* against San Jose scale was examined in three apple orchards, each located at Srinagar, Bandipora and Pulwama districts in Kashmir. The relative pathogenicity of these fungi against the pest was also evaluated during field trials. Mortality was monitored at 2-day intervals until 30 days after application. All three fungal pathogens caused mortality of the pest particularly with the increase in treatment concentration. High mortality (76-77%) was determined with *B. bassiana* at  $15 \times 10^5$  conidia/ml. formulation followed by *L. lecanii* at the same formulation (mortality 73-75%) at all the experimental sites tested. *M. anisopliae* sensu lato was significantly less effective (mortality 47-67%) in all the three field trials. The results demonstrate the suitability of entomopathogenic fungi for controlling San Jose scale.

KEY WORDS: Biological control, Quadraspidiotus perniciosus, Hemiptera, Diaspididae, entomopathogenic fungi

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# **INTRODUCTION**

Quadraspidiotus perniciosus San Jose scale (Comstock) (Hemiptera: Diaspididae) is a key pest of apple in certain hilly tracts of India (Malik et al., 1972; Masoodi et al., 1993). Its distribution throughout the temperate regions of the world and its expansion to additional host species make this insect a serious pest. Female San Jose scales produce crawlers which settle on the bark, leaves and fruit and because of their small size are difficult to detect visually. A single female produces up to 500 crawlers (Korchagin, 1987) and crawler emergence continues from middle of May to middle of October in Kashmir apple orchards (Masoodi and Trali, 1987). If crawlers from heavy infestations are left untreated, they may cause appreciable fruit damage.

Biological control based on parasites and predators have been tested with variable success (Masoodi and Trali, 1987; Rawat *et al.*, 1988; Masoodi *et al.*, 1989a, b; Thakur *et al.*, 1989; Thakur *et al.*, 1993; Masoodi *et al.*, 1996; Buhroo *et al.*, 2000). Among the causal agents of diseases in insects such as protozoans, bacteria, viruses, rickettsia and nematodes, the entomogenous fungi also play a relevant role. There are minimal effects of entomopathogenic fungi on non-targets and they offer a safer alternative for use in IPM than chemical insecticides (Goettel and Hajek, 2000; Pell *et al.*, 2001; Hajek and Delalibera, 2010; Khan *et al.*, 2012).

The aim of this study was to test the susceptibility of San Jose scale to the widely used entomopathogenic fungi - *Beauveria bassiana* (Bals.) Vuill, *Metarhizium anisopliae* sensu lato (Metsch.) Sorokin, and *Lecanicillium lecanii* (Zimm.) Zare and Gams and to study their comparative effectiveness during field application in different apple orchards.

# MATERIALS AND METHODS

# **Experimental orchards**

The trials were carried out in three apple orchards, each located at Srinagar, Bandipora and Pulwama districts in Kashmir. At each experimental site, the orchards had many apple cultivars but Red Delicious was the predominant cultivar. Each orchard was spread over 0.81 hectares having 15-20 year old trees and the rows planted at a dis-

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tance of 5 meters from each other. The average height of the trees was 3.5 meters ( $\pm$ 1.5 SD) and trees were infested with San Jose scale. The orchards were selected mainly on the basis of heavy infestation caused by the pest in these areas during the last 2-3 years. At each district, 30 infested apple trees were labeled for different applications.

#### **Fungal treatment**

The commercial formulations of insect pathogenic fungi containing Talc as dispersant were obtained from Varsha Bioscience and Technology, Vinay Nagar, Saidabad, Hyderabad - 500 059. They included Beauveria bassiana NCIM 1216 (spore count 1 × 108 CFU /g.), Metarhizium anisopliae sensu lato NCIM 1311 (CFU 1 × 108/g) and Lecanicillium lecanii NCIM 1312 (CFU  $1 \times 10^{8}/g$ ). The products were stored under cryogenic conditions. Conidial suspensions of each fungus for bioassays were made in distilled water at three concentrations - low  $(5 \times 10^5 \text{ conidia}/$ ml), medium (1  $\times$  10<sup>6</sup> conidia/ml) and high (15  $\times$  10<sup>5</sup> conidia/ml). The fungal treatments (5 litres of each formulation) were applied with the help of a foot sprayer to the complete tree. Treatments consisted of application to three replicate trees with each of the three fungi at each of 3 concentrations (low, medium and high). In the vicinity of these applications, three infested apple trees were sprayed with distilled water which served as control trees during the course of experimentation.

At each district, the treatments were given at 10 days after the emergence of first crawlers. This helped to provide the additional host material (fresh as well as old scales) to the fungal pathogen.

Live San Jose scales were counted on the surface of the bark on five, 1 cm<sup>2</sup> areas per tree (=1 replicate). The areas selected for counting were based on large insect population presence. This was done one day before treatment (one spray only) and at subsequent interval of 2-days after treatment for a period of 30 days. During counting, the waxy covers of the scales were carefully removed with the help of a scalpel. The shrunk and flaccid scales under the waxy cover were treated as dead. The percentage mortality was calculated at each experimental site.

#### Statistical analysis

Statistical analyses were performed using SPSS version 16.0 for Windows. The individual means of mortality and their Standard Error (SE) were calculated for each site and means of the treatment effects were separated using Tukey's HSD test. The treatment effects were statistically significant at  $P \le 0.05$ .

# **RESULTS AND DISCUSSION**

## **Experimental site 1**

The data collected on percentage mortality at the Hazratbal experimental site is presented in Table 1. The results revealed that the scales on infested apple trees in the orchards were highly susceptible to the three fungal species tested. At low concentration (5  $\times$  10<sup>5</sup> conidia/ml), mortality of scales reached a maximum of 60.07% (±0.69 SE) with B. bassiana, 47.39% (±1.05 SE) with M. anisopliae, and 60.97% (±0.65 SE) with L. lecanii. B. bassiana and L. lecanii were statistically similar (P = 0.502) but both produced significantly higher mortality than M. anisopliae (P = 0.000) at this concentration. At concentration formulation ( $1 \times 10^6$  conidia/ml), mortality reached a maximum of 68.66% (±0.33 SE) with B. bassiana, 56.77% (±0.33 SE) with M. anisopliae, and 64.51% (±0.55 SE) with L. lecanii, all statistically different from each other (P = 0.000). At high concentration ( $15 \times 10^5$  conidia/ml), mortality reached a maximum of 75.87% ( $\pm 0.58$  SE) with *B. bassiana*, 66.19% (±0.83 SE) with *M. anisopliae*, and 73.15% (±0.79 SE) with L. lecanii. There were no significant differences between the mortalities caused by B. bassiana and L. le*canii* at this concentration (P = 0.188) but both the species caused significantly higher mortality than M. anisopliae  $(P \le 0.005).$ 

#### **Experimental site 2**

The data collected on percentage mortality at the Bandipora experimental site is presented in Table 2. The applications again showed that the scales infesting apple trees were highly susceptible to the fungal species tested. When low concentrated treatment was applied, the mortality of scales reached a maximum of 61.19% (±0.57 SE) with B. bassiana, 53.20% (±0.92 SE) with M. anisopliae, and 61.63% (±0.62 SE) with L. lecanii. The concentrations of B. bassiana and L. lecanii were significantly at par (P = 0.302) but both produced significantly higher mortality than *M. anisopliae* ( $P \le 0.018$ ). At medium concentration, mortality reached a maximum of 69.32% (±1.26 SE) with B. bassiana, 57.08% ( $\pm 0.85$  SE) with M. anisopliae, and 65.95% (±0.92 SE) with L. lecanii. All the three fungal species were significantly different from each other at this concentration ( $P \le 0.050$ ). At high concentration, mortality reached a maximum of 76.82% (±0.29 SE) with B. bassiana, 66.67% (±1.32 SE) with M. anisopliae, and 73.34% (±0.79 SE) with L. lecanii. There were no significant differences between B. bassiana and L. lecanii at this concentration (P = 0.102) but both caused significantly higher mortality than *M. anisopliae* ( $P \le 0.050$ ).

| ortality of San Jose scale on apple trees due to entomopathogenic fungi at Hazratbal, Srinagar | Days after application |
|--|------------------------|
| umulative percentage mortali   | Concentra-             |
| Table 1. Cu  | Treatments             |

| Treatments   | Concentra-                          |               |              |                           |                 |                |          | Jays after ap | plication |          |          |          |          |          |          |
|--|-------------------------------------|---------------|--------------|---------------------------|-----------------|----------------|----------|---------------|-----------|----------|----------|----------|----------|----------|----------|
|  | tion                                | 4             | 9            | 8                         | 10              | 12             | 14       | 16            | 18        | 20       | 22       | 24       | 26       | 28       | 30       |
| B. bassiana  | Low                                 | 0.01          | 4.16         | 2.43                      | 4.51            | 10.07          | 13.19    | 20.48         | 35.42     | 43.40    | 44.09    | 51.39    | 55.55    | 60.07    | 60.07    |
|  |                                     | (3.69)a       | (4.21)a      | (1.39)a                   | (3.02)a         | (1.25)a        | (1.25)a  | (0.34)a       | (0.60)a   | (0.34)a  | (0.34)a  | (0.92)a  | (0.91)a  | (1.39)a  | (0.69)a  |
|  | Medium                              | 1.00          | 43.33        | 15.33                     | 21.66           | 26.66          | 34.33    | 39.33         | 45.66     | 52.00    | 57.33    | 63.66    | 67.33    | 68.66    | 68.66    |
|  |                                     | (6.42)a       | (3.17)a      | (2.33)a                   | (1.66)b         | (0.88)b        | (2.40)b  | (3.17)b       | (1.20)b   | (0.57)b  | (0.66)b  | (1.45)b  | (0.88)b  | (0.33)b  | (0.33)b  |
|  | High                                | 17.03         | 18.80        | 26.45                     | 35.27           | 46.45          | 53.81    | 56.45         | 58.52     | 62.63    | 66.16    | 67.93    | 70.28    | 73.81    | 75.87    |
|  |                                     | (3.18)a       | (4.04)a      | (3.11)a                   | (1.55)c         | (1.63)c        | (0.77)c  | (1.28)c       | (0.00)c   | (0.77)c  | (0.77)c  | (0.59)b  | (0.58)b  | (0.77)b  | (0.58)c  |
| M. anisopliae  | Low                                 | 1.10          | 1.45         | 8.12                      | 11.27           | 19.69          | 25.30    | 28.46         | 40.38     | 43.54    | 45.64    | 49.50    | 49.50    | 47.39    | 47.39    |
|  |                                     | (2.18)a       | (1.40)a      | (3.65)a                   | (1.26)a         | (2.29)d        | (3.03)d  | (2.78)a       | (0.35)a   | (0.35)a  | (0.35)a  | (1.21)a  | (0.00)c  | (1.05)c  | (1.05)d  |
|  | Medium                              | 3.30          | 6.93         | 9.24                      | 13.53           | 16.50          | 19.47    | 22.44         | 27.39     | 33.33    | 44.55    | 48.18    | 50.49    | 56.77    | 56.77    |
|  |                                     | (4.05)a       | (1.51)a      | (4.13)a                   | (1.74)ad        | (0.87)bd       | (2.31)e  | (1.83)d       | (0.87)d   | (2.00)d  | (0.57)ad | (0.87)a  | (0.99)cd | (0.33)d  | (0.33)e  |
|  | High                                | 6.16          | 7.74         | 10.90                     | 11.21           | 17.22          | 20.37    | 25.75         | 30.17     | 36.17    | 44.07    | 48.50    | 53.24    | 60.19    | 66.19    |
|  | I                                   | (3.79)a       | (1.92)a      | (2.50)a                   | (1.37)a         | (5.17)d        | (1.09)f  | (1.37)d       | (1.37)d   | (2.75)d  | (1.89)a  | (1.92)a  | (1.13)c  | (0.54)d  | (0.83)f  |
| L. lecanii   | Low                                 | 3.92          | 4.90         | 7.85                      | 15.40           | 22.94          | 27.52    | 34.08         | 36.71     | 46.54    | 51.47    | 53.76    | 58.68    | 60.98    | 60.97    |
|  |                                     | (1.31)a       | (6.58)a      | (1.42)a                   | (0.00)a         | (0.86)e        | (1.42)de | (4.43)a       | (2.14)a   | (1.18)a  | (0.86)c  | (0.56)a  | (0.56)a  | (0.86)a  | (0.65)a  |
|  | Medium                              | 4.39          | 9.83         | 15.91                     | 21.02           | 27.10          | 29.33    | 35.73         | 38.29     | 47.24    | 48.20    | 54.92    | 55.55    | 63.87    | 64.51    |
|  |                                     | (2.30)a       | (1.66)a      | (1.28)a                   | (1.39)ac        | (5.33)be       | (3.14)be | (0.96)ab      | (2.09)ae  | (0.55)ae | (0.55)e  | (0.55)ac | (0.84)e  | (0.32)ae | (0.55)ag |
|  | High                                | 10.41         | 14.93        | 20.96                     | 24.28           | 34.54          | 40.87    | 50.22         | 50.53     | 59.27    | 61.99    | 66.51    | 69.23    | 73.15    | 73.15    |
|  |                                     | (5.81)a       | (5.03)a      | (2.87)a                   | (3.70)ac        | (0.30)ce       | (1.20)eg | (0.90)e       | (0.30)f   | (1.56)c  | (0.52)c  | (0.52)b  | (1.04)b  | (0.60)b  | (0.79)c  |
| Distilled wa-  |                                     | 0.00          | 00.00        | 0.00                      | 0.00            | 0.00           | 0.00     | 0.00          | 0.00      | 0.00     | 0.00     | 0.00     | 0.00     | 2.44     | 3.35     |
| ter (control)  |                                     | (0.00)        | (0.00)       | (0.00)                    | (0.00)          | (0.00)         | (0.00)   | (0.00)        | (0.00)    | (0.00)   | (0.00)   | (0.00)   | (0.00)   | (0.34)   | (4.04)   |
| *Each figure is a mean of 3 replicates with SEM in parenthesis.<br>*The values followed by same letter within each column do not differ si | mean of 3 replic<br>wed by same let | cates with SE | M in parenth | tesis.<br>A not differ si | ionificantly (' | Tiikev's test) |          |               |           |          |          |          |          |          |          |

The values followed by same letter within each column do not differ significantly (Tukey's test)

Table 2. Cumulative percentage mortality of San Jose scale on apple trees due to entomopathogenic fungi at Quil, Bandipora

| Treatments   | Conc.         |               |               |                 |          |                           | _        | Days after a | upplication |        |          |         |          |          |          |
|--|---------------|---------------|---------------|-----------------|----------|---------------------------|----------|--------------|-------------|--------|----------|---------|----------|----------|----------|
|  |               | 4             | 9             | 8               | 10       | 12                        |          |              | 18          |        | 22       | 24      | 26       | 28       | 30       |
| B. bassiana  | Low           | 9.78          | 14.09         | 16.08           | 19.07    | 24.04                     | 24.70    | 26.36        | 30.68       | 39.63  | 44.94    | 46.26   | 55.55    | 61.19    | 61.19    |
|  |               | (4.23)a       | (2.89)a       | (1.32)a         | (0.87)a  | (0.66)a                   | (3.51)a  |              | (2.01)a     |        | (1.75)a  | (0.57)a | (2.32)a  | (0.57)a  | (0.57)a  |
|  | Medium        | 10.49         | 13.03         | 17.77           | 27.57    | 29.79                     | 35.16    |              | 47.50       |        | 48.45    | 52.24   | 61.73    | 64.89    | 69.32    |
|  |               | (1.75)a       | (1.76)a       | (1.37)a         | (0.63)a  | (1.09)a                   | (1.92)a  |              | (1.58)b     |        | (1.13)a  | (1.92)a | (1.38)a  | (0.54)a  | (1.26)b  |
|  | High          | 19.33         | 28.43         | 34.59           | 38.70    | 45.45                     | 51.90    |              | 64.50       |        | 71.54    | 73.89   | 75.57    | 76.82    | 76.82    |
|  | 1             | (4.60)a       | (0.77)b       | (1.78)b         | (2.79)b  | (1.83)b                   | (1.92)b  |              | (1.27)c     |        | (1.17)b  | (0.77)b | (0.29)b  | (0.29)b  | (0.29)c  |
| M. anisopliae  | Low           | 6.43          | 11.64         | 22.12           | 25.96    | 28.41                     | 35.04    |              | 42.03       |        | 43.42    | 47.61   | 50.06    | 52.15    | 53.20    |
|  |               | (2.77)a       | (1.39)a       | (1.74)c         | (1.25)c  | (0.35)c                   | (0.60)c  |              | (2.12)d     |        | (0.60)a  | (0.60)a | (0.35)a  | (0.34)c  | (0.92)d  |
|  | Medium        | 13.83         | 18.36         | 21.26           | 28.36    | 33.84                     | 39.97    |              | 47.40       |        | 47.72    | 53.85   | 55.79    | 57.08    | 57.08    |
|  |               | (2.43)a       | (3.07)a       | (1.70)ac        | (0.55)ac | (0.64)ac                  | (1.93)ac |              | (0.64)bd    | $\sim$ | (1.67)a  | (0.85)a | (0.64)ac | (0.85)cd | (0.85)de |
|  | High          | 10.23         | 16.83         | 24.42           | 26.73    | 34.65                     | 39.93    |              | 52.81       |        | 56.11    | 60.73   | 65.35    | 66.34    | 66.67    |
|  |               | (4.58)a       | (0.99)a       | (1.83)c         | (1.51)c  | (0.57)d                   | (1.43)c  |              | (0.33)e     |        | (1.43)c  | (0.66)c | (1.98)d  | (0.99)e  | (1.32)f  |
| L. lecanii   | Low           | 9.74          | 18.23         | 20.75           | 28.93    | 30.19                     | 42.76    |              | 47.17       |        | 55.03    | 56.92   | 58.17    | 62.57    | 61.63    |
|  |               | (5.29)a       | (1.91)a       | (0.54)a         | (1.57)d  | (0.54)e                   | (1.36)d  |              | (0.54)f     |        | (0.83)cd | (0.63)d | (1.25)a  | (0.83)a  | (0.62)a  |
|  | Medium        | 20.22         | 30.97         | 31.89           | 39.25    | 36.49                     | 48.46    |              | 56.74       |        | 59.19    | 63.49   | 64.10    | 65.95    | 65.95    |
|  |               | (1.33)a       | (2.43)b       | (0.53)d         | (1.06)e  | (4.63)ae                  | (0.53)e  |              | (1.06)g     |        | (1.22)e  | (1.10)e | (0.53)ae | (0.92)a  | (0.92)g  |
|  | High          | 20.03         | 32.61         | 40.70           | 44.59    | 49.68                     | 55.07    |              | 60.16       |        | 64.96    | 66.75   | 69.75    | 72.44    | 73.34    |
|  |               | (2.25)a       | (2.26)b       | (0.90)e         | (3.03)f  | f(0.89)f                  | (0.51)b  |              | (1.66)c     |        | (0.51)f  | (1.37)f | (0.30)f  | f(0.79)f | (0.79)c  |
| Distilled water  |               | 0.00          | 0.00          | 0.00            | 0.00     | 0.00                      | 0.00     |              | 0.00        |        | 0.00     | 0.00    | 3.10     | 3.81     | 0.00     |
| (control)  |               | (0.00)        | (0.00)        | (0.00)          | (0.00)   | (0.00)                    | (0.00)   |              | (0.00)      |        | (0.00)   | (0.00)  | (3.05)   | (1.33)   | (0.00)   |
| *Each figure is a mean of 3 replicates with SEM in parenthesis.          | ean of 3 repl | icates with S | EM in parent  | hesis.          |          |                           |          |              |             |        |          |         |          |          |          |
| " The values followed by similar letter within each column do not differ | ea by similar | letter withit | r each columi | an ao not aine. |          | signincanuy ( iukeys tesi | (J       |              |             |        |          |         |          |          |          |

Biocontrol efficacy of entomopathogenic fungi against San Jose scale

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| Treatments  | Conc.          |                |             |          |          |         | D       | ays after a <sub>l</sub> | pplication |          |         |         |          |          |          |
|---|----------------|----------------|-------------|----------|----------|---------|---------|--------------------------|------------|----------|---------|---------|----------|----------|----------|
|   |                | 4              | 9           | 8        | 10       | 12      | 14      | 16                       | 18         | 20       | 22      | 24      | 26       | 28       | 30       |
| B. bassiana   | Low            | 2.33           | 5.66        | 14.33    | 23.66    | 29.00   | 32.66   | 38.00                    | 43.33      | 45.66    | 51.33   | 55.33   | 57.00    | 61.66    | 61.66    |
|   |                | (5.23)a        | (2.60)a     | (3.17)a  | (2.18)a  | (1.73)a | (0.66)a | (3.21)a                  | (0.33)a    | (0.88)a  | (1.20)a | (1.20)a | (1.00)a  | (0.66)a  | (0.66)a  |
|   | Medium         | 6.07           | 9.23        | 19.67    | 25.68    | 38.33   | 48.45   | 54.46                    | 57.30      | 59.20    | 61.41   | 63.63   | 65.52    | 69.00    | 69.00    |
|   |                | (2.51)a        | (2.58)a     | (1.67)a  | (0.83)a  | (2.19)a | (1.92)b | (1.09)b                  | (1.64)b    | (0.54)b  | (0.63)b | (0.31)b | (0.83)b  | (1.67)b  | (1.14)b  |
|   | High           | 14.06          | 14.06       | 22.90    | 29.86    | 42.49   | 48.18   | 53.23                    | 56.71      | 61.45    | 68.08   | 71.56   | 73.46    | 74.09    | 77.25    |
|   |                | (0.63)a        | (3.34)a     | (1.13)a  | (1.97)a  | (0.83)b | (2.06)b | (1.37)b                  | (0.31)b    | (0.83)b  | (0.63)c | (0.94)c | (0.00)c  | (0.83)c  | (1.63)c  |
| M. anisopliae   | Low            | 2.42           | 12.15       | 22.92    | 27.43    | 31.60   | 32.98   | 36.80                    | 40.97      | 41.66    | 42.36   | 43.40   | 44.09    | 50.34    | 53.12    |
|   |                | (0.34)a        | (1.51)b     | (0.60)b  | (2.11)a  | (2.50)a | (2.27)a | (0.69)a                  | (1.25)a    | (1.04)a  | (2.11)d | p(69.0) | (0.34)d  | (0.34)d  | (1.04)d  |
|   | Medium         | 5.66           | 9.33        | 12.66    | 15.66    | 21.66   | 26.33   | 32.00                    | 39.33      | 44.00    | 57.00   | 50.00   | 52.66    | 55.66    | 56.33    |
|   |                | (4.09)a        | (4.33)ab    | (1.45)a  | (2.33)a  | (1.20)c | (0.33)c | (1.52)c                  | (1.76)c    | (1.52)ac | (1.73)b | (1.00)e | (0.66)e  | (0.66)e  | (0.33)de |
|   | High           | 15.61          | 24.02       | 35.13    | 46.84    | 51.35   | 54.95   | 56.76                    | 59.46      | 61.86    | 63.36   | 64.26   | 67.57    | 61.86    | 61.86    |
|   |                | (1.82)a        | (1.08)b     | (1.37)c  | (2.26)b  | (0.51)d | (1.04)b | d(06.0)                  | d(06.0)    | bd(07.0) | (0.60)b | (0.30)f | f(06.0)  | (0.79)e  | (0.79)ad |
| L. lecanii  | Low            | 2.07           | 11.83       | 22.98    | 29.95    | 32.74   | 36.57   | 42.50                    | 55.04      | 48.42    | 50.86   | 55.04   | 57.48    | 62.36    | 62.36    |
|   |                | (1.74)a        | (2.72)b     | (1.25)b  | (2.76)a  | (2.44)a | (2.78)a | (1.59)a                  | (1.80)d    | (1.51)e  | (0.60)a | (0.60)a | (1.25)a  | (1.59)a  | (0.60)a  |
|   | Medium         | 13.98          | 19.51       | 30.59    | 36.13    | 37.11   | 40.69   | 42.32                    | 45.25      | 46.89    | 51.12   | 54.70   | 58.94    | 63.17    | 65.78    |
|   |                | (2.03)a        | (4.59)ab    | (2.45)bd | (1.81)ac | (0.86)a | (0.32)d | (2.45)a                  | (1.12)e    | (2.28)ef | (0.56)a | (0.32)g | (0.56)dg | (0.32)af | (0.56)ab |
|   | High           | 15.54          | 24.82       | 39.50    | 39.50    | 49.08   | 55.07   | 59.56                    | 62.56      | 66.75    | 69.15   | 73.04   | 73.34    | 74.84    | 74.84    |
|   |                | (1.03)a        | (3.45)b     | (1.30)be | (1.30)ac | (1.19)b | (1.03)b | (1.37)b                  | (0.30)f    | (0.51)g  | (1.82)c | (0.51)c | (1.07)c  | (1.03)c  | (0.51)c  |
| Distilled water   |                | 0.00           | 0.00        | 0.00     | 0.00     | 0.00    | 0.00    | 0.00                     | 0.00       | 0.00     | 0.00    | 4.31    | 0.00     | 4.00     | 2.44     |
| (control)   |                | (0.00)         | (0.00)      | (0.00)   | (0.00)   | (0.00)  | (0.00)  | (0.00)                   | (0.00)     | (0.00)   | (0.00)  | (2.30)  | (0.00)   | (1.30)   | (0.34)   |
| *Each figure is a mean of 3 replicates with SEM in parenthesis. | nean of 3 repl | licates with S | EM in paren | uthesis. |          |         |         |                          |            |          |         |         |          |          |          |

Each figure is a mean of 3 replicates with SEM in parenthesis. The values followed by same letter within each column do not differ significantly (Tukey's test)

# **Experimental site 3**

The data on percentage mortality at the Awantipora experimental site is presented in Table 3. The fungal applications caused sufficient mortalities of the scales on apple trees. At low concentration, the mortality of scales reached a maximum of 61.66% (±0.66 SE) with *B. bassiana*, 53.12%  $(\pm 1.04 \text{ SE})$  with *M. anisopliae*, and 62.36%  $(\pm 0.60 \text{ SE})$ with L. lecanii. When medium concentration was applied, the mortality reached a maximum of 69.00% (±1.14 SE) with B. bassiana, 56.33% ( $\pm 0.33$  SE) with M. anisopliae, and 65.78% (±0.56 SE) with L. lecanii. At high concentration, mortality reached a maximum of 77.25% (±1.63 SE) with B. bassiana, 61.86% (±0.79 SE) with M. anisopliae, and 74.84% (±0.51 SE) with L. lecanii. The concentrations of B. bassiana and L. lecanii were not significant and produced significantly higher mortality than M. anisopliae (P  $\leq 0.002$ ) at all the concentrations.

In control trees, there was negligible mortality of San Jose scale during the experimental period. The maximum mortalities at any of the sites caused by fungal pathogens at each of the three treatment concentrations are summarized in Figure 1. The data revealed that there were no significant differences between *B. bassiana* and *L. lecanii* among the fungal species at each of the three treatment concentrations (P = 0.723 for low concentration; P = 0.127 for medium concentration; and P = 0.343 for high concentration). However, both the species produced significantly higher mortality than *M. anisopliae* at each treatment concentration ( $P \le 0.009$ ). The overall maximum mortality was obtained with *B. bassiana* at high conidial concentration ( $15 \times 10^5$  conidia/ml).

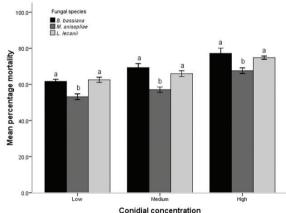


Fig. 1. Comparison of fungal bioefficacy against San Jose scale at three different concentrations. Different letters above bars (mean  $\pm$  1SD) indicate statistical significance (Tukey's test)

There is little information available on the susceptibility of San Jose scale to entomopathogenic fungi. This work demonstrates that entomopathogenic fungi are capaBiocontrol efficacy of entomopathogenic fungi against San Jose scale

ble of infecting San Jose scale and killing the early settled crawlers and nymphs on the bark of the apple tree. All three fungal pathogens used in the present study showed high efficacy against the pest especially with the increase in the concentration of the treatments. This may be due to residual effect of entomopathogenic fungi which results in release of cutin-degrading enzyme, toxin and other metabolites inimical to insect pest (St. Leger, 1995; Vey *et al.*, 2001) and also due to prophylactic action of the fungus on the scale pest. The efficacy of the pathogenic fungi was more or less similar when repeated in all three districts of the valley.

The fungal pathogen B. bassiana has been tested and developed as a commercial mycoinsecticide by a number of researchers in the USA (e.g. Bradley et al., 1992; Poprawski et al., 1999; Vandenberg et al., 1998). Finally it was allowed for commercial use in 1999 by the U.S. Environmental Protection Agency. It is a promising biocontrol candidate used on a large variety of tree and field crops for control of grasshoppers, whiteflies, thrips, aphids and many other insect pests in North America (Shah and Pell, 2003). The present results showed that among the three species of entomopathogenic fungi, the highest mortality- 77.25% was caused by *B. bassiana* at  $15 \times 10^5$  conidia /ml. concentration followed by L. lecanii (at the same concentration) at all the three experimental sites. The high mortality obtained with B. bassiana corroborate with the reports of Sheeba et al., (2001) on rice weevils where B. bassiana produced mortality up to 75.8% when monitored at 5-day intervals until 25 days. Similar, observations on B. bassiana causing maximum mortality of 71.10% in plant bug (Liu et al., 2003) and 80% in broad mite (Nugroho and Ibrahim, 2004) were reported. In addition commercial preparations of B. bassiana were infective after more than 12 months' storage at 25 °C (Wraight et al., 2001).

In the present experiments, *L. lecanii* also produced better results and caused more than 70% mortality against San Jose scale at all the three experimental sites. This pathogen has already been recommended for control of aphids and related insects in Europe (Shah and Pell, 2003) and good efficacy against a number of aphid species has been demonstrated (Hall, 1981; Milner, 1997; Burges, 2000; Yeo *et al.*, 2003).

The present investigations showed that among the three species of entomopathogenic fungi used, *M. anisopliae* was significantly less effective than the other two against San Jose scale.

The study had established an alternative for synthetic insecticides so as to formulate the ecofriendly management strategies against San Jose scale. It has been noted (Shah and Pell, 2003) that most entomopathogenic fungi are best used when total eradication of a pest is not required, but

instead insect populations are controlled below an economic threshold, with some crop damage being acceptable. Therefore, entomopathogenic fungi be used in conjunction with other conventional and cultural methods in IPM. The safety of entomopathogenic fungi towards humans, the environment and non-target organisms is clearly an important criterion for consideration and each insect-fungus system must be evaluated on a case-by-case basis.

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