AN ASSESSMENT ON POPULATION DENSITY OF SAN JOSE SCALE QUADRASPIDIOTUS PERNICIOSUS (COMSTOCK) AND ITS BIOLOGICAL CONTROL IN KASHMIR (HEMIPTERA: DIASPIDIDAE)

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ABSTRACT: San Jose scale *Quadraspidiotus perniciosus* is a key pest of apple crop in the northern states of India. An assessment on its population density was carried out in five districts of Kashmir Valley. In district Baramulla, the pooled mean scale population ranged from 10.29 per cm^2 area to 37.32 / cm^2 over the course of its active period from April to October. This population range was 10.74–36.45 scales /cm² area in district Bandipora, 11.39-37.48 /cm² area in district Srinagar, 10.22-35.57 /cm² area in district Anantnag, and 10.14-33.72 /cm² area in district Budgam. The efficacy of entomopathogenic fungi-Beauveria bassiana. Metarhizium anisopliae sensu lato and Lecanicillium lecanii at three concentrations against the pest was examined in an experimental orchard. Mortality of the pest was monitored at 2-day intervals until 30 days after application and the maximum mortality was used for data analysis. All three fungal pathogens caused mortality of the pest particularly with the increase of treatment concentration. High mortality (77%) was determined with B. bassiana at 15×10^5 conidia /ml. concentration followed by L. lecanii at the same concentration (mortality 75%). However, M. anisopliae sensu lato was significantly less effective (mortality 53-68%) among the three concentrations tested during field trial. The results demonstrate the suitability of entomopathogenic fungi for controlling San Jose scale.

KEY WORDS: Population density, *Quadraspidiotus perniciosus*, Hemiptera, Diaspididae, biological control

San Jose scale *Quadraspidiotus perniciosus* (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 & Trali, 1987; Buhroo et al., 2000). 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 & Trali, 1987; Rawat et al., 1988; Masoodi et al., 1989a,b; Thakur et al., 1989; Thakur et al., 1993; Masoodi et al., 1996). 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 & Hajek, 2000; Pell et al., 2001; Hajek & Delalibera, 2010; Khan et al., 2012).

The objective of this study was to assess the population density of San Jose scale in Kashmir and to test the effectiveness of various concentrations of entomopathogenic fungi– *Beauveria bassiana* (Bals.) Vuill, *Metarhizium anisopliae* sensu lato (Metsch.) Sorokin, and *Lecanicillium lecanii* (Zimm.) Zare & Gams against the pest during field trial.

MATERIALS AND METHODS

Population density

San Jose scale population density was assessed in five districts of Kashmir viz. Baramulla, Bandipora, Srinagar, Anantnag and Budgam during the year 2008. At each district three orchards were taken and from each orchard ten apple trees (Red Delicious cultivar) were randomly selected. Orchards were categorized as high, medium and least infested on visual basis taking into account live scale population. The twigs of selected trees were examined for recording scales per square centimeter area on five spots in each tree. The observations were recorded at fortnightly intervals from last week of March to October.

Field trial

The field trial for determining efficacy of fungal applications was carried out in an apple orchard located at Pulwama district in Kashmir. At the trial site, the orchard had many apple cultivars but Red Delicious was the predominant cultivar. The orchard was spread over 0.81 hectares having 15-20 year old trees and the rows planted at a distance 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 orchard was taken mainly on the basis of heavy infestation caused by the pest during the preceding years and 30 infested apple trees were labeled for different applications.

Fungal treatment

The commercial forms of insect pathogenic fungi were obtained from Varsha Bioscience and Technology, Vinay Nagar, Saidabad, Hyderabad-500 059. They included *Beauveria bassiana* NCIM 1216 (spore count 1×10^8 CFU /g.), Metarhizium anisopliae sensu lato NCIM 1311 (CFU 1 \times 108 /g.) and Lecanicillium lecanii NCIM 1312 (CFU 1×10^8 /g.). Each product also contained Talc as a dispersant. 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 10⁵ conidia /ml.), medium (1 \times 10⁶ conidia /ml.) and high $(15 \times 10^5 \text{ conidia /ml.})$. 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). Beauveria bassiana at low concentration was applied to three trees, medium concentration to three trees and high concentration to three trees (and the same was done for *M. anisopliae* and L. lecanii). 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 the treatment site, the treatments were started 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^2 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 of San Jose scale was calculated at the experimental site.

Statistical analysis

Statistical analyses were performed using SPSS version 20.0 for Windows. All data were analyzed using descriptive statistics and the percentage mortalities after applications were separated using Tukey's HSD test. The treatment effects were statistically significant at $P \le 0.05$.

RESULTS AND DISCUSSION

The data collected on population density of San Jose scale in district Baramulla is presented in Figure 1. The results revealed that the pooled mean of live scale population was 10.29 per cm² area at the end of March which increased to a peak of 37.32 /cm² at the end of July and from there onwards it gradually declined to 26.48 /cm² area in the first fortnight of October. The data collected in district Bandipora (Fig. 2) revealed that the pooled mean population of the scales was 10.74 /cm² area in the 1st week of April which reached to a maximum of 36.45 $/cm^{2}$ area at the end of July and thereafter slowly declined to a low of 25.45 $/cm^{2}$ area until the middle of October. In district Srinagar (Fig. 3) the live scale population was 11.39 /cm² area in the first week of April which gradually increased to a maximum of 37.48 /cm² area up to the first week of August. Then the population declined to a low of 26.32 /cm² area up to the middle of October. The data collected in district Anantnag (Fig. 4) revealed that the population of the scales was 10.22 /cm² area in the first week of April which increased to a maximum of 35.57 /cm² area in the first week of August and then it came down to 24.17 /cm² area in the third week of October. The data collected at district Budgam (Fig. 5) showed a population of 10.14 scales $/\text{cm}^2$ area in the first fortnight of April which gradually increased to 33.72 /cm² area in the first week of August and then it again declined to $24.39 \ /cm^2$ area in the second fortnight of October.

The above observations showed that the sequence of population level of San Jose scale in different apple orchards remained more or less the same throughout the districts surveyed in Kashmir. The peak population was always observed in August in all the districts surveyed. However, the maximum population was observed in districts Srinagar and Baramulla followed by districts Bandipora, Anantnag and Budgam.

The data collected on percentage mortality at the Awantipora experimental site is presented in Figure 6. The treatments showed that the scales infesting apple trees were highly susceptible to the fungal species tested and the high mortality was achieved on 30^{th} day after treatment. At low concentration (5 × 10⁵ conidia /ml.), the mortality of scales reached a maximum of 61.66% (±1.15 SD) with B. bassiana, 53.16% (±1.58 SD) with M. anisopliae, and 62.56% (±1.41 SD) with L. lecanii. At medium concentration (1×10^6 conidia /ml.), mortality reached a maximum of 69.33% (±2.19 SD) with *B. bassiana*, 57.10% (±1.47 SD) with *M.* anisopliae, and 65.93% (± 1.61 SD) with L. lecanii. At high concentration (15×10^5 conidia /ml.), mortality reached a maximum of 77.23% (±2.85 SD) with B. bassiana, 67.60% (±1.55 SD) with M. anisopliae, and 74.80% (±0.90 SD) with L. *lecanii*. The data also 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

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concentration ($P \le 0.001$ for low concentration; $P \le 0.002$ for medium concentration; and $P \le 0.009$ for high concentration). The overall maximum mortality was produced by *B. bassiana* at high conidial (15 × 10⁵ conidia /ml.) concentration.

In control trees, there was almost negligible mortality ($3.58\% \pm 0.72$ SD) of San Jose scale during the experimental period. This natural mortality occurs due to environmental factors including parasitic wasps and predators.

This work demonstrates that entomopathogenic fungi are capable 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 of treatment concentration. 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 & 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 15×10^5 conidia /ml. concentration followed by *L. lecanii* (with same concentration) during the field trial. This high mortality obtained with B. bassiana is similar to the mortality observed by Sheeba et al. (2001) in rice weevils where *B. bassiana* produced mortality up to 75.8% when monitored at 5day intervals until 25 days. In similar experiments, B. bassiana caused maximum mortality of 71.10% in plant bug (Liu et al., 2003) and 80% in broad mite (Nugroho & Ibrahim, 2004). In addition commercial preparations of B. bassiana are infective even after more than 12 months' storage at 25 °C (Wraight et al., 2001). L. lecanii also produced better results and caused more than 70% mortality of the scale pest in the present experiment. This pathogen has already been recommended for control of aphids and related insects in Europe (Shah & Pell, 2003) and good efficacy against a number of aphid species has been demonstrated (Hall, 1981; Milner, 1997; Burges, 2000; Yeo et al., 2003). It was also observed that among the three species of entomopathogenic fungi used, M. anisopliae was significantly less effective than the other two against San Jose scale.

CONCLUSION

The aim of this study was to find an alternative for synthetic insecticides so as to formulate the ecofriendly management strategies against San Jose scale. It has been noted (Shah & 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 could be used against the scale pests in conjunction with other conventional and cultural methods in IPM.

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Figure 1. Pooled mean population of San José scale on Red Delicious cultivar of apple in district Baramulla.



Figure 2. Pooled mean population of San José scale on Red Delicious cultivar of apple in district Bandipora.



Figure 3. Pooled mean population of San José scale on Red Delicious cultivar of apple in district Srinagar.

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Figure 5. Pooled mean population of San José scale on Red Delicious cultivar of apple in district Budgam.



Figure 6. Pooled mean percentage mortality of San Jose scale due to entomopathogenic fungi at three different concentrations. Different letters above bars (mean \pm 1SD) indicate statistical significance (Tukey's test).