Laboratory evaluation of *Beauveria bassiana* (Balsamo) Vuillemin against *Indarbela quadrinotata* Walker (Lepidoptera: Metarbelidae) - a key pest of *Casuarina equisetifolia* L. in Tamil Nadu

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ABSTRACT: The bark eating caterpillar, Indarbela quadrinotata Walker is a serious pest of Casuarina equisetifolia in Tamil Nadu State. Survey conducted in plantations located in different parts of the State resulted in detection of a potential strain of the biocontrol agent, Beauveria bassiana. Laboratory evaluation of B. bassiana against I. quadrinotata revealed that the fungal suspension at concentrations of 2×10^6 , 2×10^7 and 2×10^8 spores/ ml was able to kill 66.67 per cent larvae. The fungus at a concentration of 4×10^8 spores/ ml gave 100 per cent mortality of the test larvae. The LC₅₀ at 240 hours was 1.6 x 10⁶ spores/ ml and the LT₅₀ for the most effective concentration was 82.86 hours.

KEY WORDS: Beauveria bassiana, Casuarina equisetifolia, Indarbela quadrinotata

The bark caterpillar, Indarbela quadrinotata Walker is a polyphagous pest attacking several forest trees belonging to families, Bombacaceae, Euphorbiaceae, Leguminosae, Loganiaceae, Myrtaceae, Rhamnaceae and Verbenaceae and also cause damage to fruit trees and bushes (Sandhu and Khangura, 1979; Bhargava and Kumawat, 1988). I. quadrinotata is a serious pest of Casurina equisetifolia in Tamil Nadu. The larva feeds on the outer as well as inner bark of trees. The pest usually attacks Casuarina trees of 2 years and above age and the mortality of trees ranges from 3 to 5 per cent annually.

Though chemical insecticides have been tested against the bark caterpillar (Mathew, 1997; Singh and Verma, 1998), use of chemical insecticides has limitations in a forest ecosystem. There are many reports on incidence of the white muscardine fungus, *Beauveria bassiana* (Balsamo) Vuillemin on insect pests of forest trees (Arshad and Hafiz, 1983; Mohamed Ali and Sudheendrakumar, 1991; Mohamed Ali *et al.*, 1991). The efficacy of the fungus, *B. bassiana* on various forest insect pests has also been reported (Mohamed Ali *et al.*, 1991). During the course of the survey on natural enemies of *I. quadrinotata*, in *C. equisetifolia* plantations

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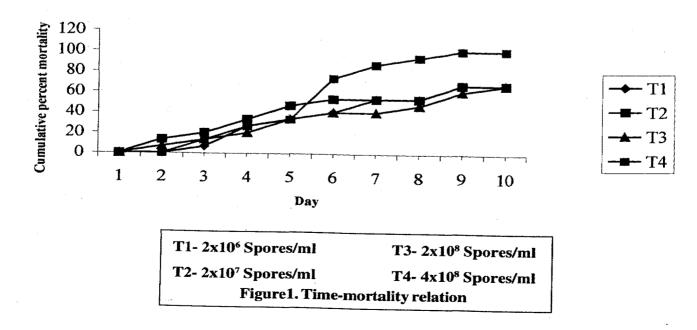
at Neyveli area of Tamil Nadu, large-scale mortality of the host larvae was found due to the infection of the fungal pathogen.

The cadavers of the bark caterpillar, *l.auadrinotata*, affected by the fungal pathogen were collected and properly washed with sterile distilled water. They were kept in Petri-dishes containing moist filter paper to facilitate fungal growth. The fungus was isolated in Potato Dextrose Agar (PDA) medium and was identified as Beauveria bassiana. Later on, cultures were made in Petri-dishes, using Saburaud Dextrose Agar (SDA) medium. The full-grown cultures after a week were crushed in sterile distilled water and filtered through sterile muslin cloth to eliminate the medium. This formed the stock spore suspension and by adding sterile distilled water made the required spore concentrations. The spore counting was made using a standard haemocytometer.

Spore suspension of *B. bassiana* in sterile distilled water, at four concentrations - 2x10⁶ spores/ml, 2x10⁷ spores/ml, 2x10⁸ spores/ ml and 4x10⁸ spores/ ml were tested for efficacy, on *I. quadrinotata*. Five bark eating caterpillars (approximately six weeks old) were introduced into Petri-dishes of 9cm diameter lined with filter paper. Fresh twigs of *Casuarina*, washed in distilled water

were provided to the larvae for feeding. Three replications, each with 5 larvae were used for the experiment. Using a pipette, 3ml of the spore suspension having the above mentioned spore concentrations were applied on to the larvae. Sterile distilled water was applied on to the larvae maintained as control. The experiment was conducted at room temperature, $27\pm1^{\circ}$ C and relative humidity 76± 4 per cent. Fresh Casuarina twigs were provided to the larvae on every third day for feeding. The larval mortality was recorded at 12 hour duration, up to 10th day. The proportion of mortality obtained at each concentration was worked out and their significance tested, using Ztest. The time-mortality relation was analyzed using Probit analysis (Finney, 1972). A software POLO-PC (© Le Ora Software, 1987), based on Finney (1972) was used for the analysis of the replicationwise data.

Out of the four concentrations -2×10^6 , 2×10^7 , 2×10^8 and 4×10^8 spores/ ml tested, the first three gave 66.67 per cent mortality of larvae by 10^{th} day after the application of spore suspension and thus, all these three concentrations were comparable in terms of larval mortality. The highest concentration tested, i. e., 4×10^8 gave 100 per cent larval mortality. The mortality at this concentration was found to be significant compared to lower concentrations



tested. No larval mortality was observed in the control. The LC_{50} values worked out at 240 hours was 1.6 x 10^6 spores/ ml. The median lethal time (LT_{50}) for the most effective dose (4 x 10^8 spores/ ml) was found to be 82.86 h.

In all the concentrations tested, the mortality of the larvae started from the 2nd or 3rd day after the inoculation (Fig.1). The infected larvae became sluggish and stopped feeding. The normal brown colour of the larvae slowly changed to pale pink. The dead larvae became somewhat stiff and mummified. Fungal mycelia were observed on the inter-segmental membranes either on 4th or 5th day after inoculation and subsequently the entire larva was covered by white, fluffy mycelial growth.

B. bassiana is highly pathogenic and has over 700 recorded host species (Moore and Prior, 1993). Rajak et al. (1993) observed that, B. bassiana caused a maximum mortality of 96 per cent to 2nd instar larvae of the teak defoliator, H. puera with a spore concentration of 1x104 conidia/ ml. They also reported that B. bassiana caused 100 per cent mortality of 2nd instar larvae of the teak skeletonizer, Eutectona machaeralis, with a spore concentration of 0.4×10^4 spores/ml. Mohamed Ali et al. (1991) studied the efficacy of B. bassiana against larvae of Ailanthus webworm (Atteva fabriciella) and found that, the fungus at concentrations of 1x10², 1x10³, 1x10⁴ and 1x10⁵ spores/ml could result in larval mortality of 26.6, 40, 53.3 and 80 per cent, respectively, at 96 hours after application. The present study has shown that a much higher spore concentration of B. bassiana is required to achieve maximum kill of the bark-eating caterpillar, I. quadrinotata.

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