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**A Study of Efficacy of fungal entomopathogens against red cotton stainer, *Dysdercus cingulatus* Fabricius (Hemiptera: Pyrrhocoridae)**

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**ABSTRACT :**

The efficacy of entomopathogenic fungi [*Beauveria bassiana* (Bb08, Bb10) and *Isaria fumosorosea* (Ifr)] isolated from different regions of Southern Tamil Nadu, India was assessed against red cotton stainer, *Dysdercus cingulatus*. All the tested isolates showed 100% mortality. The *B. bassiana* isolates such as Bb08, Bb10 and an *I. fumosorosea* isolates exemplified the significant mortality among the isolates tested. LC50, LC90 and correlation coefficient were calculated for mortality. Bb08, Bb10 and Ifr isolates unveiled the lowest LC50 ( $5.9 \times 10^5$ ,  $6.6 \times 10^5$  and  $2.6 \times 10^5$ ) and LC90 ( $1 \times 10^9$ ,  $7.3 \times 10^8$  and  $3.9 \times 10^8$ ) value compared to the isolates tested. Highly significant correlation coefficient was observed in the isolates Bb08, Bb10 and Ifr.

**Key words:** *Beauveria bassiana*, *Isaria fumosorosea*, native isolates, mortality.

Cotton, *Gossypium hirsutum* (Linn.) is the most economically important natural fiber material in the world. It is widely known as “The King of Fibers”. The economy of many countries depends up on cotton production. Nearly 24% of the total cotton production in the world is cultured from India. In recent years, cotton production is declining due to the infestation of insect pests and diseases. Of these sucking pests are deleterious during the early stage of cotton growth. Pests such as stainer, jassids, aphids, whiteflies and thrips are constituted as important pests of cotton (Uthamasamy et al., 2004). One of the dreadful pests of cotton in southeast Asian countries is *Dysdercus cingulatus* (Fab.) (Hemiptera:Pyrrhocoridae) (Kohno and Bui Thi, 2004). It is distributed in all the cotton growing regions of India (Sahayaraj and Ilayaraja, 2008). It is commonly known as red cotton

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bug and is an important pest of lady's finger, sambhal, hollyhock etc. If not managed properly, both nymph and adult insects damage the cotton bolls and leaves severely. Although it has been controlled by many synthetic insecticides because of the high residual effect of chemical insecticides, the immediate need for sustainable eco-friendly pest management has been felt very strongly providing an impetus to research and development of microbial pesticides.

Entomopathogenic fungi are widely available biological control agents (BCAs) for controlling agricultural pests (Wraight et al., 2001). Nowadays, hundreds of fungi have been identified and are being developed as biological control agents for various insects. Species from the genera *Beauveria*, *Metarhizium*, *Isaria* and *Lecanicillium* and others have been registered and commercialized in the United States. (Environmental Protection Agency, 2009). *Isaria fumosorosea* (Wize) (= *Paecilomyces fumosoroseus*) is a fungal biocontrol agent with the potential for controlling several insect pests (Dunlap et al., 2005). The species *Isaria fumosorosea* and *Isaria farinosa* are well-known entomopathogenic fungi with a worldwide distribution in temperate and tropical zones and there is a relatively wide host range which makes them interesting agents for the development of biocontrol methods. Hence, the present study aims to evaluate the potential biocontrol agent *I. fumosorosea* against *D. cingulatus*.

## **MATERIALS AND METHODS**

Entomopathogenic fungi, *Beauveria bassiana* [Bb08 (Accession No. HQ848783) and Bb10 (Accession.No. HQ416713)] and *Isaria fumosorosea* isolates used in this investigation were isolated from rhizosphere soil of different regions of Madurai and Dindigul District, Tamil Nadu and were routinely grown on potato dextrose agar (PDA). The plates were incubated at 26°C for 10-14 days and stored in a refrigerator. All the fungal isolates were sub-cultured once in three weeks. To maintain the virulence, after subculturing all the three fungal isolates were passed through host insect and re-isolated for further studies.

### **Preparation of fungal spore concentrations**

Three fungal isolates were cultured in potato dextrose agar (PDA) and were incubated at 26 °C for 10 days. After sporulation, aerial conidia were harvested by flooding the plate with sterile deionized water (dH<sub>2</sub>O) containing 0.02% Tween80. Conidial spore suspensions were prepared and conidial count determined using a haemocytometer. All the suspensions were adjusted to a concentration of 1.5 x 10<sup>8</sup> conidia mL<sup>-1</sup> from which lower concentrations were prepared by serial dilution technique for bioassay studies.

## Laboratory bioassay

Bioassays were conducted with different isolates of *B. bassiana* and *I. fumosorosea* against *D. cingulatus* adult in the laboratory with conidial suspensions containing  $1.5 \times 10^8$  conidia  $\text{mL}^{-1}$ . Twenty adult *D. cingulatus* were released on the cotton bolls sprayed with the fungal suspension and transferred into a glass jar and covered with a muslin cloth. *D. cingulatus* larvae sprayed with 0.02 % of Tween-80 were maintained as control in separate glass jars. Three replications each with 20 adults were maintained for each treatment. Mortality counts were taken at every 24 hrs after inoculation.

## Statistical Analysis

Mortality data were subjected to Probit Analysis using SPSS (version 10.0) for LC50 and LC90 prediction and Chi-Square values were calculated using Microsoft Excel.

## RESULTS AND DISCUSSION

As chemical control programmes were in practice for the management of *D. cingulatus* very few studies representing the biological control of *D. cingulatus* have been under taken so far.

**Table 1. LC50, LC90 Chi-square value and percentage of mortality caused by *Beauveria bassiana* (Bb08) against *Dysdercus cingulatus***

Time Days	LC50 (Spore $\text{mL}^{-1}$ )	LC90 (Spore $\text{mL}^{-1}$ )	Chi Square	Mortality (%)
2DAT	$5.4 \times 10^{11}$	$2.3 \times 10^{11}$	3.94* (p=0.268)	24±0.47
4DAT	$3.1 \times 10^{10}$	$1.9 \times 10^{14}$	2.976* (p=0.395)	44±0.47
6DAT	$3.3 \times 10^8$	$1.5 \times 10^{12}$	8.349* (p=0.039)	68±0.47
8DAT	$1.6 \times 10^7$	$3.3 \times 10^{10}$	2.768* (p=0.429)	88±0.47
10DAT	$5.9 \times 10^5$	$1 \times 10^9$	7.048* (p=0.070)	100±0.92

**Significant at  $p < 0.05\%$**

In the present investigation, entomopathogenic fungi isolated from rhizosphere soils were found highly virulent when tested against red cotton stainer, *D. cingulatus*. The percent mortality of the tested isolates against *D. cingulatus* was 100% at  $1 \times 10^8$  spore  $\text{mL}^{-1}$  after 10

days of post treatment accordingly. The LC50 and LC90 of the Bb08 isolate range were between  $5.9 \times 10^5$  to  $5.4 \times 10^{11}$  spore mL<sup>-1</sup> and  $1.9 \times 10^9$  to  $2.3 \times 10^{15}$  spore mL<sup>-1</sup> whereas in Bb10 and Ifr, it was  $6.6 \times 10^5$  to  $1.2 \times 10^{15}$  spore mL<sup>-1</sup> and  $7.3 \times 10^8$  to  $3.7 \times 10^{23}$  spore mL<sup>-1</sup> as well as  $2.6 \times 10^5$  to  $3.8 \times 10^{15}$  spore mL<sup>-1</sup> and  $3.9 \times 10^8$  to  $1.2 \times 10^{32}$  spore mL<sup>-1</sup> respectively. (Table 1 & 2).

**Table 2. LC50, LC90 Chi-square value and percentage of mortality caused by *Beauveria bassiana* (Bb10) against *Dysdercus cingulatus***

Time Days	LC50 (Spore mL <sup>-1</sup> )	LC90 (Spore mL <sup>-1</sup> )	Chi Square	Mortality (%)
2DAT	$1.2 \times 10^{15}$	$3.7 \times 10^{23}$	0.119* (p=0.989)	22±0.47
4DAT	$2.5 \times 10^{10}$	$8.4 \times 10^{14}$	0.827* (p=0.843)	48±0.47
6DAT	$2.9 \times 10^9$	$9.9 \times 10^{13}$	1.492* (p=0.684)	55±0.47
8DAT	$2.1 \times 10^7$	$2.3 \times 10^{10}$	1.603* (p=0.659)	88±0.74
10DAT	$6.6 \times 10^5$	$7.3 \times 10^8$	7.048* (p=0.070)	100±0.92

Among the isolates tested, Ifr had lowest LC50 ( $2.6 \times 10^5$  spore mL<sup>-1</sup>) and LC90 ( $3.9 \times 10^8$  spore mL<sup>-1</sup>) against *D. cingulatus* compared to Bb08 and Bb10 isolates, which was having LC50 and LC90 value of  $5.9 \times 10^5$ ,  $6.6 \times 10^5$  spore mL<sup>-1</sup> and  $1.9 \times 10^9$ ,  $7.3 \times 10^8$  spore mL<sup>-1</sup> respectively (Table 3). The chisquare values often represented the significant insecticidal efficiency of the tested isolates against *D. cingulatus*. Similar to the present investigation, Sahayaraj and Borgio (2010) observed 92.30% mortality of the *D. cingulatus* treated with green muscardine fungus, *Metarhizium anisopliae*. Similarly, Sahayaraj and Borgio (2010) used *M. anisopliae* for the control of *D. cingulatus* and found 75% mortality after 96 hrs of exposure. The LC50 of the present investigation was in accordance with the study reported by Sahayaraj and Borgio (2010) who observed LC50 value of  $2.25 \times 10^5$  spore mL<sup>-1</sup>. Similarly, Sahayaraj and Majesh Tomson (2010) reported the efficiency of the crude metabolites of the *M. anisopliae* capable of causing 45% mortality against *D. cingulatus*. Much in the same way, higher susceptibility of the *D. cingulatus* towards benzene extract of

*Padina pavonica* having LC50 of 0.004% was observed by Sahayaraj and Kalidas (2011). Besides, the entomopathogen is highly virulent against the caterpillar of *S. litura* (Gayathri et al., 2010; Joseph et al., 2010; Malarvannan et al., 2010; Sanehdeep et al., 2011; Suganya, and Selvanarayanan, 2010). Thus the present study exemplifies the excellent biocontrol potential of the soil isolate, *Ifr* towards red cotton stainer, *D. cingulatus*. Therefore it is recommended that isolates *Ifr* can be used as biopesticide for the control of the red cotton bug, *D. cingulatus* and other insect pests.

**Table 3. LC50, LC90 Chi-square value and percentage of mortality caused by *Isaria fumosorosea* against *Dysdercus cingulatus*.**

Time Days	LC50 (Spore mL <sup>-1</sup> )	LC90 (Spore mL <sup>-1</sup> )	Chi Square	Mortality (%)
2DAT	3.8x10 <sup>15</sup>	1.2x10 <sup>32</sup>	0.539* (p=0.910)	18±0.47
4DAT	4.8x10 <sup>11</sup>	9.5x10 <sup>16</sup>	0.678* (p=0.878)	32±0.47
6DAT	3.4x10 <sup>9</sup>	3.4x10 <sup>13</sup>	2.600* (p=0.457)	52±0.92
8DAT	1.3x10 <sup>7</sup>	5x10 <sup>10</sup>	8.210* (p=0.42)	80±0.92
10DAT	2.6x10 <sup>5</sup>	3.9x10 <sup>8</sup>	3.502* (p=0.321)	100±0.47

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