Fusarium pallidoroseum: A potential entomopathogenic agent for the biological management of Aphis gossypii

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Abstract
With rising need of switching over to sustainable agricultural practices, utilization of entomopathogenic fungi (EPF) as biocontrol agents, provides better substitute against chemical pesticides–having several side-effects. Therefore, an attempt have been made to explore the potential EPF fungi that could be incorporated into IPM practices for control of Helicoverpa armigera Hubner and Aphis gossypii Glover. Regarding this, an entomopathogenic fungus, Fusarium pallidoroseum (Cooke) Sacc, was isolated from natural population of H. armigera infesting chickpea (Cicer arietinum L.) and explored efficacy under in-vitro & field conditions. The findings of present investigation shows efficacy of F. pallidoroseum as potential biocontrol agent against okra aphid (A. gossypii), as it inflicted initial mortality of 43.33% nymphs on 2nd day and lead to complete annihilation (93.33%) of nymph population on 8th day of spore suspension application at 1×10^6 spores/mL concentration. The observations against adult okra aphid clearly demonstrated that spraying of 1 x 10^6 spores/mL of F. pallidoroseum resulted 66.67% mortality after 8th days of spraying. Increased mortality was recorded with increase in spore suspension concentrations. The LC₅₀ & LC₉₀ value for F. pallidoroseum against nymphs of A. gossypii was recorded 3.79 x 10^6 and 2.74 x 10^6, respectively. The findings were used to develop formulations (1 x 10^6 to 1 x 10^9 spore suspension/mL conc), and tested at field-level. The results showed that formulation at 1 x 10^8 spores/mL conc was most effective against A. gossypii, recorded 93.33% mortality of nymphs & 66.67% mortality of adults; could be used under IPM practices.

Keywords: Aphis gossypii, Bio-efficacy, Entomopathogen, Fusarium pallidoroseum, Helicoverpa armigera, Bioagent

INTRODUCTION: Aphis gossypii, Bio-efficacy, Entomopathogen, Fusarium pallidoroseum, Helicoverpa armigera, Bioagent

INTRODUCTION

During the last few years, H. armigera and A. gossypii are considered serious pests causing considerable losses under changing agro-climatic conditions in India, despite heavy uses of chemical pesticides (EPPO, 2006; Patel and Purohit, 2013; Ghosal et al., 2012; Khatting et al., 2016; Nagamandla et al., 2017; Singh and Dhiman, 2018; Rathee and Dalal, 2018 and Yaqoob et al., 2019). A. gossypii (Homoptera: Aphididae) is a major sucking pest of okra, causing considerable losses in the form of curling and distortion of the young leaves. Also, the presence of nymphs and adults, their shed skins and honeydews decrease the aesthetic quality of the crop. Advancement in the field of biological management of major pests of pulses, especially against the pod borer H. armigera, has been significantly in the recent past (Mehrv et al., 2008; Ahmad and Ansari 2013; Jarrahi and Safavi 2016; Mora et al., 2017; Kalvnadi et al., 2018 and Goncalves et al., 2020). Productivity of chickpea crop is greatly affected by chickpea pod borer H. armigera, which damages up to 90-95% crop because of its high fecundity, nomadic behavior, polyphagous feeding nature and induced resistance against major groups of insecticides (Mishra et al., 2013). This pod has reported the yield loss of up to 400Kg/ha- borer, with 30-40% average pods damaged during favorable environment conditions; which causes reluctant to cultivate chickpea among the farmers (Hussain, 2007). However, the continued development of natural populations resistant to chemical insecticides indicates that
further chemicals and or biological agents must be investigated for their efficacy against these insects. Considering the ill effects of chemicals and increased application costs, the biocontrol method such as use of fungal pathogens is desirable. Although, there were lots of evidence on the occurrence and efficacy of entomopathogens such as *Metarrhizium* spp and *Beauveria* spp, still there are several gaps in the identification and morphological characterization of natural enemies and their exploitation as bio-management tools with reference to agro-climatic niches. These fungal bio-control agents would offer a new approach to combat natural population of insect pest, while protecting the efforts and investments of the marginal farmers (Pawar and Borikar, 2005; Lingappa *et al*., 2005; Ahmad and Ansari 2013; Jarrahi and Safavi 2016; Kalvandi *et al*., 2018; Mohammed *et al*., 2018; Javed *et al*., 2019; Nazir *et al*. 2019 and Litwin *et al*. 2020). The indispensable step in the development of an effective microbial biocontrol agent is careful assessment and selection of the most suitable isolate, based on virulence against host insect. Factors such as temperature, pH, humidity and other environmental factors have a great significance in the incidence, severity and epidemiology of the disease (Patel and Purohit, 2013; Khating *et al*., 2016; Nagamandla *et al*., 2017; Singh and Dhiman, 2018; Rathee and Dalal, 2018 and Yaqoob *et al*., 2019). The absence of significant correlation between field and laboratory outcomes have made it complicated to visualize the genuine efficacy of entomopathogen against target and non-target insects due to different environmental conditions. Thus in the present investigation, an attempt have been made to explore an alternate entomopathogenic biocontrol agent from natural environment; which could be used as an eco-friendly, efficient, cost-effective biocontrol agent as well as can reduce the agricultural losses.

**MATERIALS AND METHODS**

**Study area**

Nineteen villages of the Tehsil Bakshi Ka Talab (BKT), district Lucknow were randomly selected for collection of the diseased *Helicoverpa* larvae. Bakshi Ka Talab is geographically located at the North Latitude 26°59’0” and East Latitude 80°53’0” E. It is situated at the distance of 25 km away from the Lucknow, at the National Highway 24. The temperature of this area was 42°C to 45°C while in winter season temperature fell down between 5°C -8°C and the elevation of BKT was 124m (407 ft). Total number of villages in this Tehsil was 185 (Fig. 1).

**Collection of infected Helicoverpa armigera larvae**

Frequent field visits were made to the selected villages of BKT, and collected the diseased specimens of *Helicoverpa* larvae. The samples thus collected were kept properly and brought to the Biocontrol Laboratory, University of Lucknow; for further identification of natural enemies.

**Isolation of entomopathogenic fungi**

The infected larvae were surface sterilized with 0.1% (w/v) mercuric chloride solution and rinsed thrice in sterile distilled water. The sterilized infected larva was placed separately into sterilized petriplates, containing potato dextrose agar with streptomycin sulphate (2.5µg/mL), for isolation of entomopathogenic fungi at 28±2°C for three days. The culture was further purified by growing single spore on PDA plates and maintained at 4°C in refrigerator.

**Effect of different media on isolated entomopathogen**

The isolated entomopathogenic fungus was further cultured on a different medium such as Czapek-Dox agar, potato dextrose agar, MYEA, agar, coconut milk agar (5 and 10 %), Martin agar medium, Molish Agar medium, Sabarouds agar medium, Richards medium and Asthana and Hawkers medium; for detailed study of morphological characteristics. A mycelial disc of 3 mm diameter was transferred and inoculated centrally onto different culture plates containing different medium in 3 replicates and incubated at 28 ± 2°C. The colony appearances and pigmentation were assessed after 2 weeks of incubation, while growth rate was measured daily until fully grown. Average dry weight of the three replicate was taken as standard value for comparing the growth in different media.

**Effect of temperature and pH on isolated pathogen**

The effect of temperature and pH was assessed by analyzing the *in vitro* growth rate on potato dextrose broth at different temperature and pH, respectively. For observing temperature effect, the PDB containing flasks were inoculated with an equal amount of fungal inoculum (20µl of test entomopathogen at the concentration 10^6 spores /mL). The flasks were incubated at different temperatures 20, 24, 28, 32, 35 and 40°C after inoculation for fifteen days. The dry weight of mycelial mats was measured 15 days after inoculation. For observing pH effect, before sterilization the pH of the medium was adjusted in the range of pH 5 to pH 9 by using 1M solution of HCl and NaOH. The flasks were inoculated with mycelial disc (3mm) of entomopathogen from the 7 days old culture. Flasks containing PDB of different pH were incubated at 28±2°C. Average dry weight of the three replicates was taken as standard value for comparing the growth of fungus at different pH.

**Scanning electron microscopy**

For SEM observations, mycelia and conidia of *F. palli-
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doroseum were fixed with 5% cold buffered glutaraldehyde for 24 h at room temperature. The samples were washed with sodium cacodylate buffer for 30 min and subsequently fixed with 2% osmium tetroxide for 24 h at 20 °C, dehydrated in a graded ethanol series for five minutes each and sputter coated with gold palladium. The images of F. pallidoroseum were obtained in the scanning electron microscope (JOEL, Japan, Model JSM 6490 LV) at the Department of Environmental Science, Babasaheb Bhimrao Ambedkar University, Lucknow. The details regarding applied voltage, magnification used and the size of the content of the images were implanted on the photographs itself.

**In vitro bio-efficacy against Helicoverpa armigera and Aphis gossypii**

Two hundred mL of culture medium was taken in a 500 mL conical flask, autoclaved at 120 °C (15 lbs) for 20 minutes. The flasks were inoculated with a six mm disc of two week’s old F. pallidoroseum grown on PDA media. The flasks were incubated at 28±2 °C in an incubator orbital shaker at 120± 10 rpm. The spore suspension was prepared by mixing the fungal mat in a mixer-grinder and mixed with 10 ml of sterile distilled water having 0.02% Tween 80 as a wetting agent (Rombach et al., 1986). The suspension was then filtered through sterile muslin cloth to eliminate the medium (Sasidharan and Varma, 2005). From the stock spore suspension, serial dilutions were made to obtain the required concentrations by using haemocytometer. Spore concentrations of 1x 10^10, 2x 10^9, 2x 10^8, 2x 10^7, 2x 10^6, 2x 10^5, 2x 10^4 and water spray as control were evaluated against H. armigera and A. gossypii, respectively (Shophiya et al., 2014 and Jayasimha et al., 2012). The lethality of the concentration was recorded by observing the percentage mortality of the okra aphid; at regular interval of two, four, six and eight days, after spraying.

**Statistical analysis**

Data on effect of temperature and pH on the growth of fungus were subjected to ANOVA, using Statistical Analysis System Version 9.0 (SAS 2002); however, for mortality data, Abbot’s formula was used to calculate corrected mortality. From the corrected mortality data, the probability integral of the chi square distribution, regression equation, slope and lethal concentrations (LC50 and LC90) were calculated; so that the efficacy and accuracy of the F. pallidoroseum can be standardized as an effective entomopathogen.

**RESULTS AND DISCUSSION**

During Rabi season; diseased Helicoverpa larvae were collected from chickpea crop of the vegetable growing areas of the study site - Bakshi Ka Talab block, Lucknow district. The samples thus collected were subjected for detailed in vitro investigation at the Bioconrol Laboratory, Department of Botany, University of Lucknow.

Out of total 1250 larvae collected from fields, 32% mortality was recorded due to tachnid flies, Campoletis chlorideae, nuclear polyhedrosis virus and fungal infection. F. pallidoroseum was isolated from third instar infected larvae of H. armigera for the first time in agro-climatic condition of Lucknow district of Uttar Pradesh, India.

**Microscopic characteristics**

The isolated fungus showed growth of dense, compact aerial mycelia, initially yellowish-orange colour and later turned white on potato dextrose medium (Fig. 2).

Fig. 1. Study area- Bakshi Ka Talab, Lucknow.
Orange to peach pigmentation around the colony on the reverse side of the Petridishes was also observed. After incubation at 28 ± 2°C temperature for 14 days, this isolate also produced definite sporodochia with macroconidia. Hyaline septate hyphae with bulged compartments, conidiophores and phialides were observed microscopically. Cylindrical phialides with small collar were observed as a constituent of a complex branching system. Monophialides as well as polyphialides conditions, were observed. Macroconidia produced in sporodochia on phialides were long, 3-9 septate, sickle shaped, smooth and possess a significant foot cell. Those born in the aerial mycelium were slightly curved, 3-7 septate and without notched. Fusiform mesoconidia were also observed in the culture which looked like ‘rabbit ears’ were abundant in the aerial mycelia.

**Scanning electron microscopy**

A big mass of hyphae was observed together with macroconidia, mesoconidia and microconidia at 1000X and 3500X magnification examined by SEM. Infection hyphae with appresorium (measuring about 5.21 µm) were observed among the mycelial mat (Fig. 3 A), along with the polyphialides (Fig. 3 B) and chlamydospore (Fig. 3 C) at 6000X and 2500X magnification, separately. The length of macroconidia was found approximately 11.26 µm (Fig. 3 D). Short spindle-shaped microconidia were also present. The mycelial organization revealed by SEM also showed an extracellular material around the hyphae which was seen as a floculent material over the cells or as a fine fibrils attaching hyphae to each other, resembling a biofilm (Fig. 3 E). The round structure seen in Fig. 3 F, under 2500X is a chlamydospore with a diameter of about 2.56 µm.

**Effect of different media**

The results on suitability of different synthetic and semisynthetic media in solid state on the growth and sporulation of the fungus are represented in Table 1. The fungus grew rapidly on solid agar medium like Czapek-Dox medium, potato dextrose medium and MYEA medium as cottony flattened colonies with yellowish-orange sporulation and produces high number of macroconidia and microconidia. Spindle-shaped mesoconidia and microconidia were observed in the aerial mycelium. The potato dextrose agar medium recorded maximum mycelial growth (80.00 mm) followed by Czapek Doxagar medium (77.66 mm) and Coconut water agar medium (10% coconut water) (75.33mm). The characteristic growth pattern of fungus was not observed on coconut agar medium with 5-10% coconut water and agar medium but production of macroconidia was found maximum.

**Effect of temperature and pH**

The optimum temperature for growth of *F. pallidoroseum* was found 28°C (dry mycelial weight 264.51 mg) followed by 32°C (dry mycelial weight 203.12 mg) and 24°C (184.43 mg). The minimum dry mycelial weight was obtained at 40°C (92.47mg) after fifteen days of inoculation which shows that higher temperature inhibited the growth of fungus (Graph 1 & Table 2).

The results revealed that the fungus could grow and sporulate in wide range of pH, i.e., from pH-5.0 to pH-9.0, in liquid potato dextrose medium (Graph 1). The dry mycelial weight was significantly higher at pH 8.0 (444.33mg) followed by pH 7 (334.42 mg) after fifteen days of inoculation. All the experimental data are significantly different from each other (Graph 2 & Table 3).

**Bio-efficacy of* F. pallidoroseum against Helicoverpa armigera**

The research results showed that *F. pallidoroseum* could initiate infection on susceptible *Helicoverpa* larvae only when the host surface was injured. This indi-
cates that this is a weak pathogen of this pest.

**Bio-efficacy of *F. pallidoroseum* against *Aphis gossypii***

Bio-efficacy studies on okra aphid, *A. gossypii* clearly indicate that this fungus might be used successfully for the control of *A. gossypii* in okra by incorporating it in integrated pest management strategies. Among the different fungal concentrations, the least per cent mortality of nymphs was noticed in $1 \times 10^6$ spore suspension (46.67%) to $1 \times 10^4$ spore suspension (36.6%) as against 93.33% in $1 \times 10^{10}$ spores per mL after 8 days after spraying (Graph 3). The result also revealed that two days after spray, all the treatments differed signifi-

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**Fig. 3.** Scan electron micrographs of *F. pallidoroseum* grown on PDA (A). Infection hyphae with appressorium (B). Polyphialides (6000X) (C). Chlamydospor (2500X) and (D). Macroconidia (3500X) (E). An extracellular material around the hyphae (F). Microconidia and chlamydospor (2500X).

cantly with each other. Nymphs were more susceptible than adults. At six days after spraying, nymphs and adults showed sluggish movement, internal and external infection and profuse sporulation. A maximum of 93.33% mortality of nymphs and 66.67% adult mortality was recorded at 1 x 10^10 spores per mL concentration which were at par with other treatments. In general, an inclination in mortality was observed with advancement of time with most spore concentrations, indicating a linear positive association between mortality and days of observation. The other treatment also resulted in significantly higher mortality of A. gossypii nymphs and adults than the control.

Bioassays of F. pallidoroseum against the nymphs and adults of A. gossypii under laboratory conditions revealed a range of variation in their biological activity. Probit analysis of mortality data enabled calculation of the dose–response relationships for nymphs, obtaining the following equations and LC_50 with 95% fiducial limits: \( y = 0.448 \pm 0.036 - 2.50 \times x^2 = 13.067 \), and \( 3.79 \times 10^5 \) spores/mL (Fiducial limit 1.84 x 10^5 – 7.12 x 10^5). The fit of the transformed data was acceptable using the chi-squared test (Table 4).

We can conclude that the isolated entomopathogen has a high potential as a biological control agent in the strategic management of A. gossypii. Thus, it can be

Table 1. Growth of Fusarium pallidoroseum on different solid media

<table>
<thead>
<tr>
<th>Medium</th>
<th>Macroconidia</th>
<th>Microconidia</th>
<th>Colour of colony</th>
<th>Radial growth after 15 days mm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Potato dextrose agar</td>
<td>++</td>
<td>+++</td>
<td>White wooly growth and orange tan in reverse</td>
<td>80.0 ± 0.171</td>
</tr>
<tr>
<td>Agar 2%</td>
<td>-</td>
<td>+++</td>
<td>Only white mycelium</td>
<td>80.0 ± 0.185</td>
</tr>
<tr>
<td>Czapek Dox Medium (Difco and BBL)</td>
<td>++</td>
<td>+++</td>
<td>Orange cousin growth with yellowish orange in reverse</td>
<td>77.66±0.168</td>
</tr>
<tr>
<td>MYEA (malt yeast extract)</td>
<td>+</td>
<td>+++</td>
<td>pink cousin growth with yellowish orange in reverse</td>
<td>72.33±0.159</td>
</tr>
<tr>
<td>Coconut water 5% with agar</td>
<td>++</td>
<td>+</td>
<td>Only white mycelium</td>
<td>67.00±0.188</td>
</tr>
<tr>
<td>Coconut water 10% with agar</td>
<td>++</td>
<td>+</td>
<td>Only white mycelium</td>
<td>75.33±0.196</td>
</tr>
<tr>
<td>Martin Agar medium</td>
<td>+</td>
<td>++</td>
<td>pink growth with yellowish orange in reverse</td>
<td>69±0.169</td>
</tr>
<tr>
<td>Molish Agar medium</td>
<td>+</td>
<td>-</td>
<td>Only white mycelium</td>
<td>22±0.086</td>
</tr>
<tr>
<td>Sabouraud Agar medium</td>
<td>+</td>
<td>+</td>
<td>Only white mycelium</td>
<td>31±0.121</td>
</tr>
<tr>
<td>Richards medium</td>
<td>++</td>
<td>-</td>
<td>Orange cousin growth with yellowish orange in reverse</td>
<td>5.8±0.001</td>
</tr>
<tr>
<td>Asthana &amp; Hawkers medium</td>
<td>++</td>
<td>++</td>
<td>White wooly growth and orange tan in reverse</td>
<td>72 ±0.173</td>
</tr>
</tbody>
</table>

Radial growth* each value is the mean of six replication and represented as mean ±SE, Sporulation represented by 0 - 20%, + 20-40%, ++ 40-60%, +++ 60-80%, ++++ 80-100%

Table 2. ANOVA for effect of different temperature on the growth of Fusarium pallidoroseum

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>SS</th>
<th>df</th>
<th>MS</th>
<th>F</th>
<th>P-value</th>
<th>F crit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between Groups</td>
<td>127648</td>
<td>7</td>
<td>18235.5</td>
<td>9.72688</td>
<td>0.00053</td>
<td>2.24902</td>
</tr>
<tr>
<td>Within Groups</td>
<td>74990.1</td>
<td>40</td>
<td>1874.75</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>202639</td>
<td>47</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 3. ANOVA for effect of media pH on the growth of Fusarium pallidoroseum.

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>SS</th>
<th>df</th>
<th>MS</th>
<th>F</th>
<th>P-value</th>
<th>F crit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between Groups</td>
<td>196392</td>
<td>7</td>
<td>28056</td>
<td>4.86266</td>
<td>0.00085</td>
<td>2.31274</td>
</tr>
<tr>
<td>Within Groups</td>
<td>186009</td>
<td>32</td>
<td>5812.79</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>382401</td>
<td>39</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
inferred that further research is needed on the possibility of developing indigenous formulation by the application of synergists or diet enhancers in increasing its virulence under field conditions. Literature reveals that the researches on *Fusarium* against insects has led to the discovery of new species (Freeman et al., 2013a; Aoki et al., 2018; Aoki et al., 2019; da Silva et al., 2020) and of numerous remarkable interactions between *Fusarium* and insects (Freeman et al., 2013b; Kasson et al., 2013; O’Donnell et al., 2016; Toki et al., 2016) demonstrating that fungi often solely studied as plant pathogens could also play supplementary roles in nature for which we don’t know the biological significance. Further, many *Fusarium* species have been recorded to be competent in controlling agricultural insect pests; causing high mortality rates and having fast action and profuse sporulation (Ganassi et al., 2001; Torres-Barragan et al., 2004; Munshi et al., 2008; Abdul-Wahid and Elbanna, 2012; Fan et al., 2015; Tosi et al., 2015; da Silva et al., 2016; Anwar et al., 2017; Velez et al., 2019; da Silva et al., 2020; Diniz et al., 2020; de Lima et al., 2021). Although, interactions of *Fusarium* spp as an entomopathogenic fungi have received greater attention in the recent years, but much remains to be explored as did in the current investigation.

### Scanning electron microscopy

During current observation of SEM studies, appressoria formation by *F. pallidoroseum* is consistent with the

### Table 4. Probit analysis of concentration-mortality response of the nymphs to *F. pallidoroseum*

<table>
<thead>
<tr>
<th>LC₅₀</th>
<th>Fiducial limit</th>
<th>LC₉₀</th>
<th>Fiducial limit</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Lower</td>
<td>Upper</td>
<td></td>
</tr>
<tr>
<td>3.79x10⁵</td>
<td>1.84x10⁵</td>
<td>7.12x10⁵</td>
<td>2.74x10⁸</td>
</tr>
<tr>
<td></td>
<td>1.13x10⁸</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Graph 1. Effect of different temperature on the growth of *F. pallidoroseum*.

### Graph 2. Effect of pH on the growth of *F. pallidoroseum*.

### Graph 3. Efficacy of *F. pallidoroseum* on the nymphs and adults of A. gossypii.
findings of Nair and Corbin (1981). Direct penetration of cuticle via infection pegs (appressoria) might also be the mode of attack, as was evident from SEM study. The observation by SEM have already been recorded against different Fusarium spp. Similar result in SEM study was recorded in case of F. solani and F. oxysporum with the difference lying in size and septation of macroconidia and microconidia facilitating taxonomic classification at the species level (Ciampi et al., 2009; Shahnazi et al., 2012; Husien, 2019).

Effect of different media
Similarly, it was revealed that potato dextrose agar medium supported the best growth of Trichoderma terrestris, Colletotrichum gloeosporioides, Beavera bassiana and F. pallidoroseum (Sheth and Ibrahim, 2014). Growth characters of F. oxysporum f. sp. gerberae studied on different solid media indicated that the growth was maximum on Oat meal agar followed by Richards’s agar, Czapek’s Dox agar, and Potato Dextrose agar supported maximum growth of fungal colony (Rajirani et al., 2000; Rajirani 2001; Chittem and Kulkarni, 2008; Mezzomo et al., 2018; Westphal et al., 2021). These studies revealed that among the solid substrates, leafy substrates, bran and oil seed cakes, and the liquid substrates tested mature coconut water supported maximum biomass and macroconidia. Increased virulence of spore suspensions prepared from coconut media was found due to the abundance of macroconida in them.

Bio-efficacy of F. pallidoroseum against Aphis gossypii
Field experiments conducted to estimate efficacy of F. pallidoroseum to manage A. craccivora using different spore formulations discovered that 82% mortality of mite, Calepitrimerus azadiractae by F. samitecum at 2.3 x 10^5 spores per mL (Navik et al., 2015). A significantly enhanced proline level in plants infected by F. pallidoroseum also showed their role as plant growth promoter (Srivastava et al., 2011). Monga et al., 2010 also reported F. pallidoroseum to cause 80-95% mortality of cotton mealybug, Phenacoccus solenopsis Tinsley. F. semitecum Berk and Ravenel was reported to be effective against many sucking pests viz., sugarcane wooly aphid, Ceratovacuca lanigera Zehntner (Aswini, 2007 and Nagaratha, 2004), on cowpea aphid, A. craccivora Koch (Roopa Rani, 2008) and tobacco aphid, Myzus persicae (Sulzer) (Asharani, 2009). Further, bio-efficacy of Fusarium pallidoroseum against cowpea sucking pests such as Aphis craccivora and Riptortus pedestris was very effective entomopathogen against A. craccivora and R. pedestris respectively and showed consistently higher mortality with increase in the exposure time; however, against Beauveria bassiana and Metarhizium anisopliae it was recorded moderately effective (Kavitha and Faizal, 2020; Singh and Kaur 2020; Tarekegn et al., 2020). Furthermore, majority of the isolates of F. oxysporum species complex have also been tested against Lepidoptera insects the mortality rates recorded from low to high (Ali-Shtayeh, and Jamous, 2003; Sun and Liu, 2008; Baidoo and Ackuaku, 2011), and from moderate to high against insects of the orders Coleoptera and Hemiptera (Torres-Barrag_an et al., 2004; Qi et al., 2011; Ameen, 2012; Qi et al., 2016; Anwar et al., 2017; Sharma and Marques 2018).

These fungal bio-control agents would offer a new approach to combat natural population of insect pest, while protecting the efforts and investments of the marginal farmers (Lingappa et al., 2005; Freeman et al., 2013; Aoki et al., 2018; Sharma and Marques , 2018; da Silva et al., 2020).

Conclusion
The findings of the present investigation shows that spraying of spore formulation of Fusarium pallidoroseum, at 1 x 10^5 spores/ mL conc was the most effective against A. gossypii, recorded 93.33% mortality of nymphs and 66.67% mortality of adults; could be used under IPM practices. Further, after multilocational field trials as well as synergistic effects of the selected strains of F. pallidoroseum, in combination with selected chemicals and their toxicity to the target pests (i.e. still in progress); an effective, ecofriendly, cost effective biocontrol agent could be explored for transferring the technology to the farmers.

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Conflict of interest
The authors declare that they have no conflict of interest.

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