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Research Article

Efficacy of Entomopathogenic Fungi against Fall Armyworm (FAW) in Laboratory Conditions

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ABSTRACT

Fall armyworm (FAW) is most dangerous pest of maize crops and distributed widely in various countries of the world especially tropical and subtropical regions, resulting threat to food security. The efficacy of Metarhizium anisopliae and Beauveria bassiana were checked against 2nd instar larvae of FAW under controlled conditions. The results showed that among tested entomopathogenic fungi, B. bassiana was found more toxic than M. anisopliae. B. bassiana caused highest larval mortality than M. anisopliae. In the current study, B. bassiana and M. anisopliae caused 68.89% and 53.85% larval mortality, respectively. M. anisopliae and B. bassiana demonstrated LT50 values of 80.54 h and 83.32 h, respectively. Additionally, the LC50 values for M. anisopliae and B. bassiana were determined to be 1.4×107 and 1.5×107 spores/ml, respectively. Entomopathogenic fungi can give effective control against pest and further studies are needed to check the efficacy against older instars under field and laboratory conditions.

Keywords: *Fall armyworm; Spodoptera frugiperda; Maize; Entomopathogenic fungi; Pakistan.*

INTRODUCTION

FAW scientifically known as *Spodoptera frugiperda*, is a member of the order Lepidoptera and the family Noctuidae. It has been documented in numerous countries across the world. It was first observed in Africa in 2016, followed by Pakistan in 2017, India in 2018, and China in 2019 (Montezano et al., 2018). Although it is native to tropical and subtropical regions globally, its presence has extended to other parts of the world. The larvae of FAW cause significant damage to various host plant species, specifically those belonging to the families Asteraceae, Fabaceae, and Poaceae (Shylesha et al., 2018; Day et al., 2017; Guo et al., 2018; Goergen et al., 2016; & Ramanujam et al., 2020).

Larvae of the FAW are an invasive insect species, pose a significant threat to various important crops such as rice, sorghum, cabbage, cotton, wheat, tomato, millet, beet, groundnut, onion, potato, and soybean.

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Maize, scientifically known as *Zea mays* and commonly referred to as the "queen of cereals," is a crucial crop worldwide due to its high reproductive capacity and its role as a source of livestock feed. However, it is highly susceptible to infestation by FAW (Romero-Arenas et al., 2014; & Opisa et al., 2018). The larvae primarily target the tender parts of plants, including young plant parts and even the cob itself. This relentless attack has resulted in annual maize yield losses of 8-20 million tons in Africa. FAW is increasingly becoming a major threat to maize production in Pakistan, and due to its ability to feed on multiple host plants, it is spreading to other crops as well (Cruz-Avalos et al., 2019; Ahmad et al., 2021; & Kumela et al., 2019).

Hence, urgent measures need to be taken to control the existing pest at an early stage in order to minimize crop yield losses. Various management strategies have been implemented worldwide to combat insect pests, including the use of chemical, biological, and botanical approaches (Ramzan et al., 2019; & Murtaza et al., 2019). Chemicals, such as insecticides, offer a prompt and effective response against insect pests but come with negative consequences for the environment, natural predators, and can lead to the development of insecticide resistance. In Pakistan, it is crucial to develop environmentally friendly and safe pest management practices that minimize insecticide resistance and environmental pollution (Wu et al., 2019; & Ramzan et al., 2020). Entomopathogenic microorganisms, including fungi, nematodes, bacteria, and viruses, offer a sustainable and harmless solution for controlling insect pests. These microorganisms have proven to be effective against the larvae of FAW, but no specific studies related to entomopathogenic fungi have been conducted against this notorious agricultural pest in Pakistan. Therefore, the objective of the present study was to evaluate the toxicity of entomopathogenic fungi against the larvae of FAW under controlled laboratory conditions.

MATERIALS AND METHODS

Second instar FAW larvae were obtained from various unsprayed maize fields in district Multan. The larvae, along with maize leaves, were transferred to the Rearing Laboratory located at the Institute of Plant Protection. In the laboratory, the larvae were kept under specific conditions, including a temperature of 26.2°C, relative humidity of 75%, and a 14:10 h day-light photoperiod. To assess their effectiveness, two entomopathogenic fungi, namely *Beauveria bassiana*, and *Metarhizium anisopliae* were tested against the second instar larvae of FAW. Spore concentrations of various levels (1×10^4, 1×10^5, 1×10^6, 1×10^7, and 1×10^8 spores/ml) were prepared using Neubauer's improved hemocytometer. Second instar larvae from the second generation, which were of equal age and size, were collected from the culture and utilized for the present study. Each replication consisted of three sets, with 10 larvae per set, which were immersed in a suspension of 1×10^8 spores/ml for a duration of 20 seconds. These treated larvae were then transferred to petri dishes containing clean and equally sized fresh maize leaves as their food source. The larvae were provided with new and fresh maize leaves daily for feeding. The control group was treated with distilled water only, without any additional substances. The mortality responses resulting from fungal infection were observed, recorded, and noted in terms of dose and time. The percentage of larval mortality was determined using Abbott's formula (Abbott, 1925). The collected data were subjected to statistical analysis using one-way analysis of variance (ANOVA) in the Statistical Package for the Social Sciences (SPSS) software, specifically the Windows version 20.0.

RESULTS AND DISCUSSION

The fall armyworm (FAW) poses a significant threat to agricultural crops, particularly maize, and is considered the most destructive pest in many regions. As an invasive species, it has become a major concern for food security in Pakistan. The prevalent practice among farmers in the study area is the extensive use

of chemical insecticides for controlling this emerging pest, which leads to environmental pollution and harm to beneficial organisms such as predators and parasitoids. Considering the negative impacts of insecticides, there is a growing need for eco-friendly and alternative approaches to manage insect pests, including FAW (Ramzan et al., 2021a; & Abid et al., 2021). Microbial control, involving the use of fungi, bacteria, viruses, and protozoa, has emerged as a viable alternative method (Akutse et al., 2019; & Safder et al., 2023). The larvae of FAW are susceptible to these microorganisms, making microbial control an important component of integrated pest management strategies. Numerous studies conducted in different countries have reported the isolation of fungi from various stages of FAW, such as eggs, larvae, pupae, and adults, and investigated their infectivity or toxicity. However, such research studies focusing on FAW and its interaction with fungi are lacking in Pakistan. It was observed that *B. bassiana* was most toxic fungus as compared to *M. anisopliae.* In the current study, *B. bassiana* and *M. anisopliae* caused 68.89% and 53.85% larval mortality, respectively (Table 1). The current study findings are almost similar to other studies conducted by many researchers on FAW in both laboratory and field conditions (Ramzan et al., 2021a).

Table 1: Percentage mortality of FAW larvae by the use of fungi

Isolates	%age mortality
B. bassiana	68.89± 0.23[a]
M. anisopliae	53.85±0.45[bc]
Control	0.76±0.45[c]
CD@1%	0.428

M. anisopliae and *B. bassiana* strains had showed 78.6 and 96.6% larval mortality at 1 × 109 conidia/ml. Our findings are different to many previous researchers. They had reported that *M. anisopliae* isolates cause 97% mortality while 30% with *B. bassiana* while *B. bassiana* showed maximum pathogenicity as compared to *M. anisopliae* in the current study. Our current study findings are different from others researchers may be due to variation in larval genetic makeup. The current and previous results variations are due to isolation of fungus strains from different sources. The pathogenicity of microbial agents can vary according to agricultural practices and geographical location. Some researchers had reported 97-100% larval mortality infected with entomopathogenic fungi (Bohnenblust et al., 2016; & Cock et al., 2017). It has already discussed that entomopathogenic fungi show high mortality at early instars and high dose as given in table 2. During the study, only 0.76% larval mortality was recorded in control.

Table 2: Dose mortality response of entomopathogenic fungi against *Spodoptera frugiperda* larvae

Isolates	LC_{50} spores/ml	95% fiducial limit	Slope±SE	$\chi 2$	P value	df
B. bassiana	1.5×10^{7}	7.02×10^{4}–7.5×10^{7}	0.382±0.199	1.854	0.492	2
M. anisopliae	1.4×10^{7}	5.51×10^{9}–4.7×10^{7}	0.496±0.206	1.531	0.597	2

In the experiment, the entomopathogenic fungi *M. anisopliae* and *B. bassiana* demonstrated LT50 values of 80.54 h and 83.32 h, respectively. Additionally, the LC50 values for *M. anisopliae* and *B. bassiana* were determined to be 1.4×107 and 1.5×107 spores/ml, respectively, as depicted in Tables 3. When applied to newly hatched FAW larvae, *B. bassiana* exhibited a mortality rate of 19% and an LC_{50} range of 7.4 × 104 conidia−1. On the other hand, the *M. anisopliae* isolate CP-MA1 demonstrated a higher mortality rate of 72.5% with an LC50 of 5.3 × 105 conidia/ml. Cruz-Avalos et al. (2019) and García et al./ (2011) reported the similar findings about larval mortality.

Table 3: Time mortality response of entomopathogenic fungi against *Spodoptera frugiperda* larvae

Isolates	LT_{50} hours	95% fiducial limit	Slope±SE	$\chi 2$	P value	df
M. anisopliae	80.54 h	79.00–174.93	7.01±1.25	1.40	0.478	2
B. bassiana	83.32 h	74.56–200.01	8.45±1.32	1.83	0.501	2

CONCLUSION

In the current study, the toxicity of entomopathogenic fungi against larvae at different instar stages was evaluated. Each concentration of the fungi exhibited a significant impact on *S. frugiperda* larvae. As the concentration of the fungi increased, the mortality rate of the larvae also increased. These findings hold great importance for future studies and provide valuable insights for controlling pest populations in both laboratory and field conditions.

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Conflict of interest

Authors announce no conflict of interest in this paper.

Author Contribution:

All authors have participated in critically revising of the entire manuscript and approval of the final manuscript.

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