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Biological control of *Spodoptera frugiperda* J. E. Smith and *Schistocerca piceifrons piceifrons* Walker using entomopathogenic fungi

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ABSTRACT

Objective: Evaluate the biological effectiveness of native entomopathogenic fungi to control *Spodoptera frugiperda* and *Schistocerca piceifrons piceifrons*, as well as the natural incidence of parasitoids in *S. frugiperda*.

Design/Methodology/Approach: Six strains of *Metarhizium anisopliae* (A1, A2, A3, A4, A5, and A6) and three strains of *Isaria fumosorosea* (B4, B5, and B6) were collected. *S. frugiperda* larvae were inoculated with all the isolated strains of *Metarhizium* and *Isaria*; while, the nymphs of *S. p. piceifrons* were inoculated with strains A1, A5, A6, B4, and B5. Mortality (%) and natural incidence of parasitoids in *S. frugiperda* larvae were evaluated. Mortality was analyzed using a one-way ANOVA and a comparison of means (Duncan; $\alpha=0.05$) in the INFostat 2021 software. Parasitism was reported with descriptive statistics (%).

Results: Strains A1, A6, and B6 caused the highest mortality (86.6-90.0 %) in *S. frugiperda* larvae. Strains A1, A5, A6, and B6 caused the highest mortality in nymphs of *S. p. piceifrons* (90-100%). Two families of parasitoids were recorded: Tachinidae (Diptera; 7.8 %) and Braconidae (Hymenoptera). Wasps of the genus *Meteorus* sp. account for 92.2% of the latter family.

Study Limitations/Implications: The biological effectiveness evaluations of the entomopathogenic fungi were carried out under laboratory conditions. The results must still be validated on the field.

Findings/Conclusions: Strains A1, A5, and A6 showed a good control of the *S. p. piceifrons* nymphs. Strain B6 is a biological control alternative for *S. p. piceifrons* and *S. frugiperda*, since it recorded the highest mortality for both species.

Keywords: Fall armyworm, Central American Locust, control alternative, *Meteorus*.

Citation: Torres-Acosta, R. I., Felipe-Victoriano, M., Garay-Martínez, J. R., & Torres-de los Santos, R. (2023). Biological control of *Spodoptera frugiperda* J.E. Smith and *Schistocerca piceifrons piceifrons* Walker using entomopathogenic fungi. *Agro Productividad*. <https://doi.org/10.32854/agrop.v16i12.2763>

Academic Editors: Jorge Cadena Iniguez and Lucero del Mar Ruiz Posadas

Received: August 17, 2023.

Accepted: November 23, 2023.

Published on-line: January 31, 2024.

Agro Productividad, 16(12) suplemento. December. 2023. pp: 13-23.

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INTRODUCTION

The fall armyworm [*Spodoptera frugiperda* J.E. Smith (Lepidoptera: Noctuidae)] and the Central America locust [*Schistocerca piceifrons piceifrons* Walker (Orthoptera: Acrididae)] are key pests that attack cotton (*Gossypium hirsutum* L.), rice (*Oryza sativa* L.), sugarcane (*Saccharum officinarum* L.), corn (*Zea mays* L.), sorghum (*Sorghum vulgare* Moench), and soybean (*Glycine max* L.) (Fotso-Kuate *et al.*, 2019; Overton *et al.*, 2021). In tropical regions, they can cause of up to 100% losses in production; consequently, in several African and

Central American countries, they are considered a threat to food security (Barrientos-Lozano *et al.*, 2021; Servín and Mendoza, 2022).

S. frugiperda is native to Central America and it is widely known throughout the continent (Jing *et al.*, 2021). In recent years, it has become an invasive pest in Africa, Asia, and Australia (Paredes-Sánchez *et al.*, 2021), where it is considered corn's main pest. It has a great number of hosts, a wide migratory capacity, high fertility, and a rapid development of resistance against chemical and biological pesticides, such as *Bacillus thuringiensis* Berliner (Jing *et al.*, 2021; De Souza-Ribas *et al.*, 2022). Just in twelve producing countries in Africa, grain yield losses range from 8.3 to 20.6 million of tons per year, which accounts for 2.5-6.2 billion of dollars in economic losses (Day, 2017). *S. p. piceifrons* has two stages: the solitary stage, in which it does not damage crops, and the gregarious stage, in which they group in swarms of millions of individuals. Their great migratory capacity allows them to move from one country to another, causing economic losses, both in agriculture and cattle raising (Le Gall *et al.*, 2019; Pérez-Ramírez *et al.*, 2019; Barrientos-Lozano *et al.*, 2021).

S. frugiperda and *S. p. piceifrons* control is mainly based on chemical management, given the high efficiency of molecules and the availability of the products. Depending on the pest species that needs to be controlled, pesticides are made up of mixed or individual formulations using active ingredients from the following chemical families: avermectins, benzoylphenyl ureas, carbamates, diamides, spinosyns, phenylpyrazoles, neonicotinoids, organophosphate, and pyrethroids (Paredes-Sánchez *et al.*, 2021; Kulye *et al.*, 2021; Birkhan *et al.*, 2023).

Currently, there are different environmentally friendly alternatives for pest control, including crops Bt (*Bacillus thuringiensis*), natural enemies (parasitoids and predators) and entomopathogenic fungi. Gutiérrez-Ramírez *et al.* (2015) and Ordóñez-García *et al.* (2015) point out that *S. frugiperda* can be attacked by parasitoids from various families of the orders Hymenoptera (Braconidae, Ichneumonidae, Platygasteridae, and Trichogrammatidae) and Diptera (Tachinidae), whose parasitism rate ranges from 3 to 42%. In this regard, local parasitoids from these families have already been reported in Mexico and in several African countries; consequently, these parasitoids can help to keep this pest under control in the Old World (Koffi *et al.*, 2020). Regarding entomopathogenic fungi, *Metarhizium anisopliae* (Mechnikov) Sorokin can cause a 43-100% mortality in third-instar larvae of *S. frugiperda* (Ullah *et al.*, 2022b; Munywoki *et al.*, 2022) and a 70-100% mortality in nymphae and adults of *S. p. piceifrons*. Additionally, it can cause mortality in others species of the Acrididae family, including *Melanoplus sanguinipes* Fabricius (Barrientos-Lozano *et al.*, 2021; Dakhel *et al.*, 2019). Genus *Metarhizium* is widespread in natural ecosystems and can be used (along with *Beauveria bassiana* (Bals.-Criv.) Vuill) to control the Central America locust (Brunner-Mendoza *et al.*, 2019; Barrientos-Lozano *et al.*, 2021).

Altinok *et al.* (2019) and Gandarilla-Pacheco *et al.* (2021) have reported that *Isaria fumosorosea* (Wize) Kepler, B. Shrestha & Spatafora is efficient to control Lepidoptera. This species can cause a 29-100% mortality among *S. frugiperda* and *S. litura* Fabricius larvae (Lei *et al.*, 2020; Ullah *et al.*, 2022a). However, there are no reports about the effect of *I. fumosorosea* on *S. p. piceifrons*. Consequently, in view of the high economic cost and the irreparable environmental damage caused against beneficial fauna, soils, and water

bodies by the excessive application of the pesticides used to control *S. frugiperda* and *S. p. piceifrons*, the objective of this study was to evaluate the biological efficiency of native entomopathogenic fungi for the control of the said species, as well as the natural incidence of parasitoids on *S. frugiperda*. The results will help to implement efficient and accessible control measures that will minimize the risks posed to human health and the environment.

MATERIALS AND METHODS

The research was carried out in 2022, in the Laboratorio de Toxicología of the Campo Experimental Las Huastecas (CEHUAS), Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP); located at 22° 33' 58.56" N and 98° 9' 49.91" W, in the municipality of Altamira, Tamaulipas, México.

Origin of the entomopathogenic fungi

The entomopathogenic fungi strains of *M. anisopliae* e *I. fumosorosea* were collected from the soils of the González, Llera, and Mante Tamaulipas municipalities, in June 2022 (Table 1). They were isolated using the insect tramp technique (Zimmermann, 1986, modified by Sánchez-Peña, 2011). The isolated strains were purified through the direct transfer of conidia from larvae with mycosis to a Potato Dextrose Agar (PDA) culture medium (Hayek *et al.*, 2012). Conidia production was carried out in solid substrate (commercial rice), at the Laboratorio de Botánica of the Unidad Académica Multidisciplinaria Mante (UAMM), Universidad Autónoma de Tamaulipas. The Humber taxonomic keys were used to identify the microscopic (conidia, phialides, and hyphae) and macroscopic (growth and color) morphology of the entomopathogenic fungi. The strains were preserved at the UAMM lab, using the inclined tube technique with mineral oil (Sharma and Smith, 1999).

Biological effectiveness test of *Metarhizium anisopliae* and *Isaria fumosorosea* for the control of *Spodoptera frugiperda*

Three-hundred *S. frugiperda* larvae in their 4th and 5th instars were collected from *Urochloa fasciculata* (Sw.) R. Webster grass, in an experimental plot of the CEHUAS. In the Laboratorio de Toxicología, the larvae were kept in an expanded polystyrene icebox until the evaluation was carried out. To prevent cannibalism, they were fed abundant grass.

To conduct the biological effectiveness test, the *S. frugiperda* larvae were inoculated with six *M. anisopliae* strains (A1, A2, A3, A4, A5, and A6) (Table 1), three *I. fumosorosea* strains (B4, B5, and B6), and a control treatment (distilled water + Tween 20 at 0.03%). The conidia suspensions were carried out in a distilled water solution plus Tween 20 at 0.03% and with conidia produced in 12 g of rice as solid production medium.

The conidia suspension of the entomopathogenic fungi was adjusted to a 1.5×10^8 and a 5.1×10^8 conidia/mL concentrations for the *M. anisopliae* and *I. fumosorosea* strains, respectively. These concentrations were standardized according to *Metarhizium* A2 and *Isaria* B5, the entomopathogenic fungi strains that produced the lowest number of conidia in 12 g of rice. Groups of 30 larvae of *S. frugiperda* were inoculated for five seconds, using the immersion technique; subsequently, they were placed in absorbent paper to remove the conidia suspension excess. Each of the *M. anisopliae* and *I. fumosorosea* strains were

Table 1. Geographic data of the entomopathogenic fungi used in the bioassays.

Entomopathogenic fungus	Code	Ejido or community	Municipality	Geographical coordinates
<i>Metarhizium anisopliae</i>	A1	Ejido El Ébano	Llera	23° 53' 98,99" N 99° 03' 44,25" O
<i>Metarhizium anisopliae</i>	A2	Rancho Nuevo del Norte	Llera	23° 41' 12,15" N 98° 98' 37,34" O
<i>Metarhizium anisopliae</i>	A3	Mante	Mante	22° 71' 65,67" N 98° 96' 24,33" O
<i>Metarhizium anisopliae</i>	A4	González	González	22° 81' 64,01" N 98° 43' 15,37" O
<i>Metarhizium anisopliae</i>	A5	El Chaparral	González	22° 51' 46,57" N 98° 44' 45,42" O
<i>Metarhizium anisopliae</i>	A6	González	González	22° 68' 47,13" N 98° 52' 77,15" O
<i>Isaria fumosorosea</i>	B4	González	González	22° 81' 64,01" N 98° 43' 15,37" O
<i>Isaria fumosorosea</i>	B5	El Chaparral	González	22° 51' 46,57" N 98° 44' 45,42" O
<i>Isaria fumosorosea</i>	B6	González	González	22° 41' 05,00" N 98° 31' 39,80" O

evaluated separately. After the inoculation of the entomopathogenic fungi, the larvae were placed separately in 30 mL plastic containers. They were provided five *U. fasciculata* leaves as a food source; the leaves were changed every 24 h. The treatments were incubated for five days, at 27 ± 1 °C and with a 85% relative humidity. Each treatment consisted of five replicates of six larvae each. Mortality was evaluated every 24 h; a larva was considered dead when it did not respond to the stimulus of a brush. Mortality was analyzed using a one-way ANOVA and Duncan's multiple range test ($\alpha=0.05$) was used to compare the means, in the INFOSTAT 2021 software (Di Rienzo *et al.*, 2020).

Natural incidence of parasitoids on the fall armyworm (*Spodoptera frugiperda*)

The emergence of parasitoids during the evaluation of the biological effectiveness of the entomopathogenic fungi against *S. frugiperda* led to the decision of evaluating natural incidence and identify the species. One-hundred thirty-three larvae from the 4th and 5th instars (size: 1-2 cm) were collected for this experiment. The larvae were placed in an icebox with plenty of grass and, subsequently, they were taken to the Laboratorio de Toxicología of CEHUAS. In the laboratory, larvae were placed individual in 30 mL plastic containers, with five *U. fasciculata* leaves (10 cm²) as food source; the leaves were changed every 24 h. The natural parasitism was evaluated for 15 days, while the number of pupae and the emergence of the parasitoids were recorded every 24 h. The first 30 parasitoids that emerged were placed in 96% ethanol, in 2 mL Eppendorf tubes; they were subsequently identified using the taxonomic keys described by Wharton *et al.* (1997). The natural parasitism of *S. frugiperda* was reported using descriptive statistics (parasitism percentage).

Biological effectiveness test of *Metarhizium anisopliae* and *Isaria fumosorosea* for the control of Central America locust (*Schistocerca piceifrons piceifrons*)

Three hundred nymphae of Central America locust (*S. p. piceifrons*) (size: 10-18 mm) were collected from the *U. fasciculata* grass of an experimental plot at the CEHUAS. The locust nymphae were taken to the Laboratorio de Toxicología of CEHUAS in an icebox. Their antennal segments were counted to determine the nymphal instar, using a Motic stereoscopic microscope. The results established that the nymphs were in their 2th and 3th nymphal instars (20-22 antennal segments).

Three strains of *M. anisopliae* (A1, A5, and A6), two of *I. fumosorosea* (B4 and B5), and a control treatment (distilled water plus Tween 20 at 0.03%) were evaluated. The conidia suspension was similar to the experiment carried out with *S. frugiperda*. The conidia concentration was adjusted to 5×10^8 conidia/mL. Groups of 50 Central America locust nymphae were separately inoculated, using the spraying technique; seven 0.3 mL sprayings were carried out with an atomizer, 20 cm away from the nymphae. Subsequently, groups of 10 specimen were placed in a 250 mL container. They were provided 20 *U. fasciculata* leaves as a food source; these leaves were changed every 24 h. The treatments were incubated for four days, at a temperature of 27 ± 1 °C and with an 85% relative humidity. Five replicates with 10 nymphae each were evaluated. Mortality was evaluated every 24 h, during four days. A nymphae was considered dead when it did not respond to the stimulus of a brush. The biological effectiveness for the control of *S. frugiperda* and *S. p. piceifrons* were carried out under laboratory conditions. The experiments were carried out using a completely randomized design, with five replicates, consisting of groups of 10 *S. p. piceifrons* nymphae per treatment. Mortality was analyzed similar to the bioassay carried out for *S. frugiperda*.

RESULTS AND DISCUSSION

Biological effectiveness test of *Metarhizium anisopliae* and *Isaria fumosorosea* for the control of *Spodoptera frugiperda*

The mortality of the *S. frugiperda* larvae caused by *M. anisopliae* recorded significant differences between treatments, 48 to 120 h after the application of the treatment ($p < 0.05$). *M. anisopliae* caused 10-20% mortality, 48 h after the start of the treatment. From 72 to 120 h after the application of the treatment, *Metarhizium* A1 and A6 caused the highest mortality, reaching 90 % (Table 2). Meanwhile, the *Metarhizium* strains A2-A5 caused the lowest mortality: 63.3 to 86.6% during the same evaluation period (Table 2).

Under laboratory conditions, *M. anisopliae* applications with a 1×10^8 conidia/mL concentration can cause a 72-100% mortality in 3rd instar larvae of *S. frugiperda* (Ullah *et al.*, 2022b). These results match the findings of this study: a 73-90% mortality among *S. frugiperda* larvae of the 4th and 5th instars can be obtained, depending on the *M. anisopliae* native strain applied. Native strains induce natural epizootics or high rates of insect mortality, as a result of the high population of pests and the high pathogenic and virulence of the entomopathogenic fungi, as well as the regional climatic conditions to which this type of fungi is adapted (Dufau *et al.*, 2021). Munywoki *et al.* (2022) and other authors have reported that a 1×10^9 conidia/mL concentration of *M. anisopliae* cause a low mortality (43%); the above is possibly due to the fact that the strain used for that study was not

appropriate for Lepidoptera. Some populations of genus *Metarhizium* can affect specific types of insects, such as *S. p. piceifrons* and *Locusta migratoria manilensis* Meyen (Brunner-Mendoza *et al.*, 2019; Barrientos-Lozano *et al.*, 2021).

Regarding *I. fumosorosea*, significant statistical differences were recorded in the mortality of *S. frugiperda* larvae between treatments, from 72-120 h after the application ($p < 0.001$). The strain B6 of *I. fumosorosea* induced the highest mortality percentages: it can cause an 86.6% and 100% mortality, 72 h and 120 h, respectively, after the application (Table 2). Meanwhile, strain *Isaria* B4 recorded the lowest mortality (76.6%), 120 h after the application of a 5.1×10^8 conidia/mL concentration.

I. fumosorosea is widely used to control Hemiptera; however, several authors have pointed out the high efficiency of this species for the control of *S. frugiperda*, *Spodoptera littoralis* Boisduval, and *Spodoptera exigua* Hübner (Altinok *et al.*, 2019; Gandarilla-Pacheco *et al.*, 2021). Therefore, it can be a biological alternative for the control of this type of pests.

Currently, there is scarce information about the effectiveness of *I. fumosorosea* against *S. frugiperda*. In this regard, Lei *et al.* (2020) pointed out that a 1×10^9 conidia/mL concentration of *I. fumosorosea* can cause 50-100% mortality in larvae of 1st-4th instars. In that study, the highest mortality rates were found in the lowest larvae instars of *S. frugiperda*. Several studies have reported that a 1×10^7 conidia/mL concentration of *I. fumosorosea* can cause a 20-50% mortality rate among the larvae of *Spodoptera litura*. Additionally, the effectiveness increased during the early larvae instars (Ullah *et al.*, 2022a).

Table 2. Biological effectiveness test of entomopathogenic fungi used for the control of fall armyworm (*Spodoptera frugiperda*).

Treatments	Mortality (%)				
	24	48	72	96	120
	Time after application (h)				
Control	0.0	6.6 ab	10.0 b	23.3 c	33.3 c
<i>Metarhizium</i> A1	0.0	0.0 b	73.2 a	86.6 a	90.0 a
<i>Metarhizium</i> A2	0.0	20.0 a	56.6 a	73.3 ab	73.3 ab
<i>Metarhizium</i> A3	0.0	0.0 b	23.3 b	49.9 b	63.3 b
<i>Metarhizium</i> A4	0.0	20.0 a	56.3 a	73.3 ab	83.3 ab
<i>Metarhizium</i> A5	0.0	10.0 ab	60.0 a	83.3 a	86.6 ab
<i>Metarhizium</i> A6	0.0	20.0 a	66.6 a	80.0 a	90.0 a
P-Value	-	0.0445	<0.0001	0.0001	0.0001
Control	0.0	6.6 a	10.0 c	23.3 c	33.3 c
<i>Isaria</i> B4	0.0	23.3 a	56.6 b	63.3 b	76.6 b
<i>Isaria</i> B5	0.0	26.6 a	69.9 ab	76.6 ab	90.0 ab
<i>Isaria</i> B6	0.0	26.6 a	86.6 a	93.3 a	100.0 a
P-Value	-	0.1908	<0.0001	0.0001	<0.0001

Different letters (a, b, c) within each species indicate a significant statistical difference between treatments (Duncan; $\alpha = 0.05$).

According to the biological effectiveness test, *M. anisopliae* and *I. fumosorosea* did not have an impact on the emergence of local parasitoids. From the 48 h to the 120 h after the application, a high percentage of parasitized larvae was observed (Figure 1A), from which parasitoid larvae emerged, which pupated and these were observed attached to a type of pedicel attached to the leaves of *U. fasciculata* or to the plastic container. The 33.6% of the 300 larvae used for the biological effectiveness test of the entomopathogenic fungi were parasitized. The emergence of adult parasitoids was observed after 5 or 6 days in the laboratory. The parasitoids were identified as wasps from the genus *Meteorus* (Braconidae). The species of this genus are polyphagous endoparasitoids of the larvae of several Coleoptera and Lepidoptera, and have been widely reported in the literature (Fujie *et al.*, 2019). Aguirre *et al.* (2015) mentioned that genus *Meteorus* prefers to parasitize larvae of the Arctiinae, Megalopygidae, Noctuidae, Nymphalidae, and Pyralidae families. These results match the findings of this study, where it parasitized larvae of *S. frugiperda* (Noctuidae).

Natural incidence of parasitoids of the fall armyworm (*Spodoptera frugiperda*)

Out of the 133 larvae collected, 48.4% (n=64 parasitoids) showed parasitism 192 h after the collection. Parasitism increased from 12.0 to 48.4%, in proportion to the incubation period of the larvae in the laboratory (Figure 1B). Of the 64 parasitoids found, two families were identified: Tachinidae (Diptera) (7.81%, n=5), of which only the pupae were observed because the parasitoids did not emerge, and wasps of family Braconidae (Hymenoptera) of the genus *Meteorus* (92.2%, n=59). According to Gutiérrez-Ramírez *et al.* (2015), Ordóñez-García *et al.* (2015), and Serrano-Domínguez *et al.* (2019), in Mexico, *S. frugiperda* presents a high parasitism rate (22-42%) by several species of Hymenoptera (Braconidae, Cheloniinae, Eulophidae, Ichneumonidae, Platygasteridae, Trichogrammatidae, and Rogadinae) and Diptera (Tachinidae). These findings match the results of this study. In the Mexican states of Durango, Sinaloa, Michoacán, and Tamaulipas, the parasitism of *S. frugiperda* by *Meteorus* ranges from 3 to 22% (Villegas-Mendoza *et al.*, 2015).

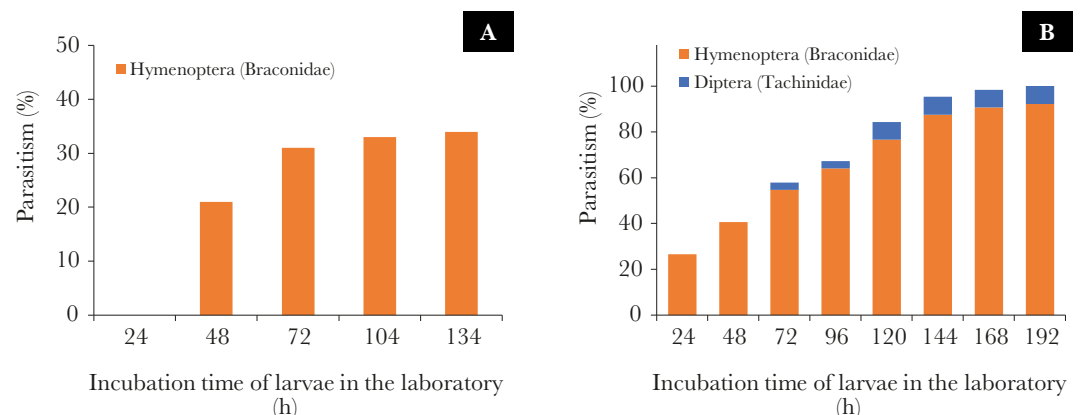


Figure 1. Incidence of natural parasitism on *Spodoptera frugiperda* larvae, with (A) and without (B) entomopathogenic fungi applications in Altamira, Tamaulipas.

In this study, the *Meteorus* sp. larvae that emerged from *S. frugiperda* caused a round wound (generally in the eighth abdominal segment), while Tachinidae caused a similar wound on the 4th and 5th abdominal segment. Both wounds had a white color that turned darker two hours after the emergence of the parasitoid. Likewise, it was observed that the larvae of *S. frugiperda* can survive 24 h after the emergence of the parasitoid larvae. This result matches the findings of Villegas-Mendoza *et al.* (2015). The formation process of the pupae of the parasitoid was not observed; however, Villegas-Mendoza *et al.* (2015) pointed out that this stage can take 40 minutes. Overall, the pupae of the parasitoid started to emerge from 24 h up to 168 h after the collection. In addition, the adults emerged from the pupae at 6.3 ± 1.5 days in the laboratory. In this regard, Villegas-Mendoza *et al.* (2015) reported that *Meteorus* sp. can emerge from the pupae, after 7.2-7.5 days at 24 ± 1 °C. Perhaps the wasps of this study emerged 1.05 days sooner than the number of days reported by these authors, because the incubation process of this research was carried out at 27 ± 1 °C.

Biological effectiveness test of *Metarhizium anisopliae* and *Isaria fumosorosea* for the control of Central American locust (*Schistocerca piceifrons piceifrons*)

Significant statistical differences between treatments were reported for the mortality of *S. p. piceifrons* nymphae caused by *M. anisopliae* and *I. fumosorosea*, 24 to 96 h after the application ($p < 0.05$) a 5×10^8 conidia/mL concentration (Table 3). *Metarhizium* strains caused a higher mortality among the nymphae of Central America locust respect to *I. fumosorosea*, perhaps as a result of the closeness of the *M. anisopliae* strain used in this research to *M. anisopliae* var. *acridum* (a very specific species which is widely used to control locust in different countries) (Kamga *et al.*, 2022). Brunner-Mendoza *et al.* (2019) and Barrientos-Lozano *et al.* (2021) have reported that *Metarhizium* sp. is an entomopathogenic fungi used to control highly specific pests. This phenomenon was also observed in this research: the three *Metarhizium* strains under evaluation caused a 98-100% mortality among Central America locusts, during the 4-day long experiment.

Table 3. Biological effectiveness tests of *Metarhizium anisopliae* and *Isaria fumosorosea* for the control of Central America locust (*Schistocerca piceifrons piceifrons*).

Treatments	Mortality (%)			
	24	48	72	96
	Time after application (h)			
Control	0.0 b	2.0 C	4.0 b	6.0 c
<i>Metarhizium</i> A1	22.0 a	92.0 a	100.0 a	100.0 a
<i>Metarhizium</i> A5	0.0 a	68.0 ab	98.0 a	100.0 a
<i>Metarhizium</i> A6	8.0 ab	70.0 ab	90.0 a	98.0 a
<i>Isaria</i> B4	4.0 b	12.0 c	12.0 b	22.0 b
<i>Isaria</i> B6	14.0 ab	56.0 b	88.0 a	96.0 a
P-Value	0.0478	0.0001	<.0001	<.0001

Different letters (a, b, c) indicate a significant statistical difference between treatments (Duncan; $\alpha = 0.05$).

At 24 h, the highest number of dead nymphs was observed in the *Metarhizium* A1 treatment, compared to the other treatments evaluated. From 72 to 96 h, the *Metarhizium* strains A1, A5, and A6 caused the highest mortality (90-100%); additionally, in statistical terms, the three local strains can cause a similar mortality. *Metarhizium* species are widely used to control pests of the Cercopidae and Acrididae families. This entomopathogenic fungi genus is very abundant in natural ecosystems and has a higher effectiveness than other entomopathogenic fungi (Brunner-Mendoza *et al.*, 2019). Currently, *Metarhizium* is the entomopathogenic fungi most used to control Central America locust and it is still subject of study, given its very specific action range against certain pests. Additionally, it can be easily reproduced (Barrientos-Lozano *et al.*, 2021). In field evaluations, *M. anisopliae* var. *acridum* caused a mortality of up to 90% among *S. p. piceifrons* adults in southeastern Mexico (Barrientos-Lozano *et al.*, 2021). Dakhel *et al.* (2019), report to *M. anisopliae* caused a 77% mortality among *Melanoplus sanguinipes* (Orthoptera: Acrididae), under greenhouse conditions.

Regarding *I. fumosorosea*, the strain B6 reached its peak mortality (96%) among *S. p. piceifrons* nymphae at 96 h, while the B4 population only reached a 22% mortality, in the same evaluation period. There are no reports about the effect of *I. fumosorosea* on *S. p. piceifrons*; this entomopathogenic fungus is generally associated with Hemiptera and Lepidoptera pests (Zimmermann, 2008; Sani *et al.*, 2023).

CONCLUSIONS

M. anisopliae strains A1, A5, and A6 showed good control of *S. p. piceifrons* nymphs under laboratory conditions in a short period, under the concentration used. Regarding *I. fumosorosea*, strain B6 represents a biological alternative for *S. p. piceifrons* and *S. frugiperda* because they caused because a high mortality in both species.

M. anisopliae and *I. fumosorosea* did not affect the emergence of local parasitoids of the Braconidae family, because a high presence of parasitism was observed under laboratory conditions, likewise the genus *Meteorus* can be considered and studied in future works to be used as a biological controller of *S. frugiperda* larvae in southern Tamaulipas.

ACKNOWLEDGEMENTS

The authors would like to thank the Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), for allowing them to carry out their research in the facilities of the C.E. Las Huastecas. Additionally, the authors would like to thank the interns of the Tecnológico de Huejutla, Hidalgo, for their help in the experiments.

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