Brazilian Journal of Biology®

ISSN 1519-6984 (Print)
ISSN 1678-4375 (Online)

Original Article

Feasibility and applications of entomopathogenic fungi in soybean cultivation with a focus on sustainability

Viabilidade e aplicações de fungos entomopatogênicos no cultivo da soja com foco na sustentabilidade

E. S. Loureiro[a,b*], G. B. Barbosa Junior[a], L. G. A. Pessoa[a], A. S. Rodrigues[a], P. M. Dias[b], M. B. Iora[a], W. K. A. Silva[a], N. E. S. Mesa[b] and M. S. Vieira[a]

[a]Universidade Federal de Mato Grosso do Sul, Faculdade de Agronomia, Chapadão do Sul, MS, Brasil

[b]Universidade Federal da Grande Dourados – UFGD, Faculdade de Ciências Biológicas e Ambientais – FCBA, Dourados, MS, Brasil

Abstract

The objective of this study was to evaluate the pathogenicity of the entomopathogenic fungus *Beauveria bassiana* (Bals.-Criv.) Vuill. on *Chrysodeixis includens* larvae in soybean cultivation. The biological insecticide Boveril® (*B. bassiana* isolate PL63, 50 g kg^{-1}) was tested against *Chrysodeixis includens* larvae, with applications at 9, 6, and 3 days before fungicide application (BA) and at 3, 6, and 9 days after fungicide application (AA). The fungicides used were Fox Xpro®, Sphere Max®, Orkestra®, and Vessarya®. A randomized block design was used, with 13 treatments and three replications: untreated control without application (T1); Turbo® pyrethroid insecticide (T2); Boveril® (positive BA control) (T3); Boveril® + Fox Xpro® BA (T4); Boveril® + Sphere Max® BA (T5); Boveril® + Orkestra® BA (T6); Boveril® + Vessarya® BA (T7); Turbo® AA (T8); Boveril® (positive AA control) (T9); Boveril® + Fox Xpro® AA (T10); Boveril® + Sphere Max® AA (T11); Boveril® + Orkestra® AA (T12); and Boveril® + Vessarya® AA (T13). The number of live *C. includens larvae* per treatment was recorded every 7 days. The data were subjected to analysis of variance and means were compared using the Scott-Knott test at $p \leq 0.05$. No negative effects of the fungicides or application intervals on the pathogenicity of *B. bassiana* against *C. includens* were detected, as the mean numbers of live larvae were not higher than those in the untreated control.

Keywords: soybean, integrated pest management, *Chrysodeixis includens*, *Beauveria bassiana*, disease management, fungicides.

Resumo

O objetivo deste estudo foi avaliar a patogenicidade do fungo entomopatogênico *Beauveria bassiana* (Bals.-Criv.) Vuill. sobre lagartas de *Chrysodeixis includens* no cultivo da soja. O inseticida biológico Boveril® (*B. bassiana* isolado PL63, 50 g kg^{-1}) foi testado contra lagartas de *C. includens*, com aplicações aos 9, 6 e 3 dias antes da aplicação de fungicidas (BA) e aos 3, 6 e 9 dias após aplicação de fungicidas (AA). Os fungicidas utilizados foram Fox Xpro®, Sphere Max®, Orkestra® e Vessarya®. Foi utilizado o delineamento experimental de blocos ao acaso, com 13 tratamentos e três repetições: controle sem aplicação (T1); inseticida piretroide Turbo® (T2); Boveril® (controle positivo BA) (T3); Boveril® + Fox Xpro® BA (T4); Boveril® + Sphere Max® BA (T5); Boveril® + Orkestra® BA (T6); Boveril® + Vessarya® BA (T7); Turbo® AA (T8); Boveril® (controle positivo AA) (T9); Boveril® + Fox Xpro® AA (T10); Boveril® + Sphere Max® AA (T11); Boveril® + Orkestra® AA (T12); e Boveril® + Vessarya® AA (T13). O número de lagartas vivas de *C. includens* por tratamento foi registrado a cada 7 dias. Os dados foram submetidos à análise de variância e as médias comparadas pelo teste de Scott-Knott ($p \leq 0{,}05$). Não foram detectados efeitos negativos dos fungicidas ou dos intervalos de aplicação sobre a patogenicidade de *B. bassiana* contra *C. includens*, uma vez que os números médios de lagartas vivas não foram superiores aos do controle sem aplicação.

Palavras-chave: soja, manejo integrado de pragas, *Chrysodeixis includens*, *Beauveria bassiana*, manejo de doenças, fungicidas.

1. Introduction

The lepidopteran pest complex is among the primary groups of soybean pests, with associated damage ranging from partial defoliation of the leaf lamina and veins to complete plant destruction (Bueno et al., 2023). Larvae of *Chrysodeixis includens* (Walker, 1858) (Lepidoptera: Noctuidae) feed on leaves in the middle and lower canopy layers, and population peaks usually occur at soybean canopy closure, which increases control complexity because

*e-mail: elisangela.loureiro@ufms.br
Received: March 23, 2026 – Accepted: April 29, 2026
Editor: Takako Matsumura Tundisi

the larvae exhibit highly aggressive feeding habits that significantly reduce the photosynthetic area of the plants (Borges et al., 2021). These population peaks occur because the larvae inhabit the lower canopy layer, where insecticides have limited penetration (Gonçalves et al., 2024).

Phytosanitary management of the crop incurs significant production costs. Integrated pest management (IPM) integrates multiple control strategies and promotes biological balance in the agroecosystem (Padilha et al., 2025). This approach is not limited to a single tool but instead relies on the integration of multiple available strategies, including natural biological control by agents present in the agroecosystem (Seni, 2023; Wang et al., 2026).

Among these important natural regulatory agents are microorganisms such as entomopathogenic fungi, which exhibit a broad spectrum of activity against arthropods and can cause epizootics and enzootics (Loureiro et al., 2024). Fungi belonging to the genera *Beauveria* (Ascomycota: Cordycipitaceae), *Metarhizium* (Ascomycota: Clavicipitaceae), and *Cordyceps* (=*Isaria*) (Ascomycota: Clavicipitaceae), are the most commonly used in applied biological control of various arthropod pests (Velozo et al., 2025).

Modern agricultural cropping systems have adopted integrated pest and disease management as a successful model because the incorrect and excessive use of pesticides, without consideration of compatibility with natural enemies, has reduced populations of these control agents and selected for resistant pest populations (Padilha et al., 2025). Phytosanitary products that are incompatible with entomopathogenic fungi can interfere with fungal spore germination and vegetative growth and may promote genetic mutations that affect fungal virulence (Loureiro et al., 2025).

Prioritizing the use of selective products compatible with entomopathogenic fungi is important for maintaining the biological balance of the agroecosystem and ensuring the effective performance of the applied phytosanitary products (Mascarin et al., 2025). This strategy offers greater safety, efficacy, and feasibility for pest and disease management (Loureiro et al., 2025), while optimizing costs associated with inputs and applications through the conservation of natural enemies in the field and adherence to the use of appropriate application intervals between fungicides and entomopathogenic fungi (Loureiro et al., 2002; Faria et al., 2023; Filgueiras et al., 2025). Studies on the toxicity of phytosanitary products to the entomopathogenic fungus *B. bassiana* support the development of new management strategies (Zambolim, 2022; Oliveira et al., 2025).

The present study evaluated a strategy to mitigate the effects of fungitoxic products on entomopathogenic fungi by interspersing applications of the fungus and fungicides at intervals that do not compromise the pathogenic performance of the fungus. The guiding question of this study concerned the optimal interval for fungicide application that would not interfere with the pathogenic activity of the entomopathogenic fungus against the target pest. The objective of this study was to evaluate the pathogenic activity of the biological insecticide Boveril® (*B. bassiana* isolate PL63, 50 g kg^{-1}) against *C. includens* larvae in soybean crops at different application intervals before and after fungicide application.

2. Material and Methods

2.1. Experimental area and management practices

The experiment was conducted in the experimental area of the Federal University of Mato Grosso do Sul (UFMS), in Chapadão do Sul, MS, Brazil (18°46′19.97″S, 52°37′05.74″W). Seeds of the soybean cultivar P98Y90 RR were sown on 2 December 2024 under a no-tillage system, with a row spacing of 50 cm and a density of 280,000 plants ha^{-1}.

Phytosanitary management was performed according to technical recommendations for soybean cultivation in the state of Mato Grosso do Sul (Embrapa, 2000). Weed control consisted of a foliar application of 2.0 kg ha^{-1} of glyphosate (Roundup® WG) plus 1.0 L ha^{-1} of manganese (Starter Manganese®) at 15 days after emergence. Soil fertilizers applied at sowing included 350 kg ha^{-1} of the 02-20-20 N–P–K formulation. The microbiological insecticide, chemical insecticide, and fungicides used in the experiment are described in Table 1. All insecticidal and fungicidal products used were applied at the medium rates recommended by the manufacturers.

Table 1. Details of the microbiological insecticide, chemical insecticide, and fungicides used in the experiment (MAPA/AGROFIT, 2025).

Commercial Product	Active ingredients	Chemical group	Formulation	Rate L ha^{-1}	Class
Boveril®	*Beauveria bassiana* (Min. 1 × 10^{8} viable conidia g^{-1})	-	WG	0.3	MI
Turbo®	Beta-cyfluthrin	Pyrethroid	EC	0.5	CI
Fox Xpro®	Bixafem; Prothioconazole; Trifloxystrobin	Carboxamide, Triazolinthione and Strobilurin	SC	0.5	CF
Sphere Max®	Trifloxystrobin; Cyproconazole	Strobilurin and Triazole	SC	0.2	CF
Orkestra®	Pyraclostrobin; Fluxapyroxad	Strobilurin and Carboxamide	SC	0.35	CF
Vessarya®	Picoxystrobin; Benzovindiflupyr	Strobilurin and Pyrazole Carboxamide	EC	0.6	CF

Rate = recommended rate by the manufacturers; MI = microbiological insecticide; CI = chemical insecticide; CF = chemical fungicide; WG = Water Dispersible Granules; EC = Emulsifiable Concentrate; SC = Suspension Concentrate.

2.2. Experimental design and treatments

The biological insecticide Boveril® (*B. bassiana* isolate PL63, 50 g kg^{-1}) was tested against *Chrysodeixis includens* larvae, with applications at 9, 6, and 3 days before fungicide application (BA) and at 3, 6, and 9 days after fungicide application (AA).

A randomized block design with 13 treatments and three replications was used. The treatments consisted of an untreated control (no applications) (T1) and applications of the following products: Turbo® pyrethroid insecticide (T2); Boveril® (positive control before fungicide application) (T3); Boveril® + Fox Xpro® BA (T4); Boveril® + Sphere Max® BA (T5); Boveril® + Orkestra® BA (T6); Boveril® + Vessarya® BA (T7); Turbo® AA (T8); Boveril® (positive control after fungicide application) (T9); Boveril® + Fox Xpro® AA (T10); Boveril® + Sphere Max® AA (T11); Boveril® + Orkestra® AA (T12); and Boveril® + Vessarya® AA (T13) (Table 2). Each plot measured 3 × 6 m (six rows spaced 50 cm apart) and was separated from adjacent plots by a 1 m uncultivated buffer to avoid border effects. Four meters of the four central rows were used for evaluations.

2.3. Applications and evaluations

Boveril® applications were performed after 4:00 p.m. under temperatures ranging from 23 to 27 °C, using a CO_2-pressurized sprayer equipped with an XR11015 nozzle, at a spray volume of 200 L ha^{-1} and a working pressure of 1.5 bar.

Fungicide applications were performed at the estágio vegetativo (V8) soybean phenological stage (45 days after planting), V8 + 14 (59 days after planting), and V8 + 28 (73 days after planting) to simulate standard fungicide applications required for soybean rust management. The same equipment described for Boveril® was used.

Monitoring of *Chrysodeixis includens* larvae began after plant emergence, with samples collected every 7 days using the beat cloth method. A 1.0 × 1.0 m cloth was placed at two points in the interrows of each plot for larval collection. The number of live *C. includens* larvae was recorded throughout the soybean cycle. Larvae were classified as small (neonate to 3rd instar, 1–10 mm) or large (4th to 5th instar, 11–25 mm).

Table 2. Description of treatments.

T1	Untreated control (free of products)
T2	Turbo® (pyrethroid) before fungicide application (BA)
T3	Boveril® (*B. bassiana*) (positive control BA)
T4	Boveril®, before the application of Fox Xpro® (BA)
T5	Boveril®, before applying Sphere Max® (BA)
T6	Boveril®, before the application of Orkestra® (BA)
T7	Boveril®, before the application of Vessarya® (BA)
T8	Turbo® (pyrethroid) after fungicide application (AA)
T9	Boveril® (*B. bassiana*) (positive control for AA)
T10	Boveril®, after application of Fox Xpro® (AA)
T11	Boveril®, after application of Sphere Max® (AA)
T12	Boveril®, after application of Orkestra® (AA)
T13	Boveril®, after the application of Vessarya® (AA)

2.4. Statistical analysis

The data were subjected to analysis of variance (ANOVA), and means were compared using the Scott-Knott test ($p \leq 0.05$) with Rbio software (Bhering, 2017).

3. Results

In the first evaluation, significant differences were detected among treatments and application intervals for the density and size of *C. includens* larvae (Table 3). At 9 days before fungicide application (BA), small larvae occurred at statistically similar densities in the Turbo®, Boveril® (positive control), and untreated control treatments. At 9 days after fungicide application (AA), treatments with Boveril® did not differ significantly (Table 3).

The occurrence of large larvae at 9 days BA did not follow the same pattern as that of small larvae. Treatments with Fox Xpro®, Sphere Max®, and Orkestra® resulted in numbers of live large larvae similar to those in the control, differing significantly from the Turbo®, Boveril®, and Vessarya® treatments, which showed lower larval occurrence (Table 3).

At 6 days BA and AA, treatments differed in the numbers of small and large larvae, except for Boveril® + Vessarya®, which showed similar mean numbers of small larvae (Table 3). At 3 days BA and AA, treatments also differed in the numbers of small and large larvae, with means statistically equal to the untreated control for Boveril® + Orkestra® (large larvae; BA), Turbo® (small and large larvae; AA), Boveril® + Orkestra® (small larvae; AA), and Boveril® + Vessarya® (large larvae; AA) (Table 3).

In the second evaluation, the occurrence of small and large *C. includens* larvae was also recorded. No significant differences were detected among treatments or application intervals (3, 6, and 9 days; BA and AA) for small larvae. The number of large larvae was significantly lower in all treatments at all intervals than in the untreated control (Table 4). In the third and fourth evaluations, no significant differences were detected among treatments, regardless of application interval or larval size (Tables 5 and 6).

In the fifth evaluation, significant differences were detected among treatments for the number of large larvae. Significant differences were also detected for small larvae across all application intervals, with the greatest population reductions observed at the 9-day intervals (BA and AA) (Table 7). At the 9-day interval BA, the Turbo® and Boveril® + Fox Xpro® treatments resulted in mean numbers of live small larvae of 3.00 and 4.33, respectively, which differed significantly from the untreated control (6.33). At the 9-day interval AA, the mean numbers of small larvae in the Turbo® (4.00), Boveril® + Sphere Max® (1.33), and Boveril® + Vessarya® (3.33) treatments were significantly different from the control (6.33) (Table 7).

At the 6-day interval BA, significant differences were detected; the Turbo®, Boveril® + Vessarya®, and Boveril® + Orkestra® treatments showed the greatest reductions in larval populations. At the 6-day interval AA, the Turbo®,

Table 3. Mean number (mean ± standard error) of live *Chrysodeixis includens* larvae observed in the first evaluation, after application of the treatments in soybean, harvest (2024/2025), Chapadão do Sul, MS, Brazil.

Treatments	Small larvae	Large larvae	Small larvae	Large larvae	Small larvae	Large larvae
	9 days		6 days		3 days	
T1 - Control	3.33 ± 0.88 a	1.67 ± 0.33 a	3.33 ± 0.88a	1.67 ± 0.33 a	3.33 ± 0.88a	1.67 ± 0.33 a
First application (BA) [3]	-	-	-	-	-	-
T2 - Turbo® (BA)	2.00 ± 0.58 a	0.67 ± 0.67 b	0.00 ± 0.00b	1.00 ± 0.58a	0.67 ± 0.67b	0.00 ± 0.00 b
T3 - Boveril® (BA)	1.33 ± 0.67 a	0.67 ± 0.33 b	1.33 ± 0.67a	0.00 ± 0.00 b	0.67 ± 0.67b	0.33 ± 0.33b
T4 - Boveril® + Fox Xpro®	0.67 ± 0.33 b	3.00 ± 0.58 a	0.33 ± 0.33b	0.00 ± 0.00 b	1.00 ± 0.58b	0.33 ± 0.33b
T5 - Boveril® + Sphere Max®	0.33 ± 0.33 b	1.67 ± 1.20 a	0.00 ± 0.00b	0.00 ± 0.00 b	0.67 ± 0.60b	0.00 ± 0.00 b
T6 - Boveril® + Orkestra®	1.00 ± 0.58 b	1.33 ± 0.67 a	0.00 ± 0.00b	0.67 ± 0.33 b	0.33 ± 0.33b	1.00 ± 0.58 a
T7 - Boveril® + Vessarya®	0.33 ± 0.33 b	0.67 ± 0.67 b	0.00 ± 0.00b	1.00 ± 0.58a	0.33 ± 0.33b	0.00 ± 0.00 b
Second application (AA)[4]	-	-	-	-	-	-
T8 - Turbo®	0.00 ± 0.00 b	0.33 ± 0.33 b	1.33 ± 0.67 a	0.00 ± 0.00 b	1.67 ± 1.20a	1.00 ± 0.58 a
T9 - Boveril®	1.00 ± 0.58 b	1.00 ± 0.58 a	0.00 ± 0.00b	0.00 ± 0.00 b	1.00 ± 0.58b	0.33 ± 0.33b
T10 - Boveril® + Fox Xpro®	1.33 ± 0.88 a	0.00 ± 0.00 b	0.00 ± 0.00b	0.00 ± 0.00 b	0.00 ± 0.00b	0.00 ± 0.00 b
T11 - Boveril® + Sphere Max®	1.67 ± 0.88 a	0.67 ± 0.33 b	0.33 ± 0.33b	0.33 ± 0.33 b	0.00 ± 0.00b	0.33 ± 0.33b
T12 - Boveril® + Orkestra®	0.67 ± 0.33 b	1.33 ± 0.67 a	1.00 ± 0.58b	1.00 ± 0.58a	2.67 ± 1.76a	0.00 ± 0.00 b
T13 - Boveril® + Vessarya®	0.33 ± 0.33 b	1.00 ± 0.00 a	2.33 ± 0.67a	1.00 ± 0.00 a	0.33 ± 0.33b	1.00 ± 0.58 a
F (treatments)	1.89*	2.24*	1.89*	2.24*	1.89*	2.24*

Means followed by equal letters within a column do not differ statistically by the Scott-Knott test at 5% probability. Larval classification: Small larvae = neonate to 3rd instar, 1–10 mm; Large larvae = 4th to 5th instar, 11–25 mm; BA = applications at 9, 6, and 3 days before fungicide application; AA = application at 3, 6, and 9 days after fungicide application (AA).

Table 4. Mean number (mean ± standard error) of live *Chrysodeixis includens* larvae observed in the second evaluation, after application of the treatments in soybean crop (2024/2025), Chapadão do Sul, MS, Brazil.

Treatments	Small larvae	Large larvae	Small larvae	Large larvae	Small larvae	Large larvae
	9 days		6 days		3 days	
T1 - Control	1.33 ± 0.88 a	5.33 ± 2.96 a	1.33 ± 0.88 a	5.33 ± 2.96 a	1.33 ± 0.88 a	5.33 ± 2.96 a
First application (BA)[3]	-	-	-	-	-	-
T2 - Turbo® (BA)	0.67 ± 0.67 a	0.33 ± 0.33 b	1.33 ± 0.33 a	0.67 ± 0.33 b	0.67 ± 0.33 a	1.00 ± 1.00 b
T3 - Boveril® (BA)	0.00 ± 0.00 a	0.67 ± 0.33 b	2.00 ± 1.15 a	2.00 ± 1.00 b	1.33 ± 0.33 a	1.33 ± 0.67 b
T4 - Boveril® + Fox Xpro®	0.33 ± 0.33 a	2.00 ± 0.00 b	0.67 ± 0.33 a	0.33 ± 0.33 b	1.00 ± 1.00 a	1.00 ± 0.58 b
T5 - Boveril® + Sphere Max®	1.00 ± 0.58 a	0.67 ± 0.33 b	1.67 ± 0.33 a	1.67 ± 1.20 b	2.00 ± 0.00 a	1.33 ± 0.67 b
T6 - Boveril® + Orkestra®	0.67 ± 0.67 a	1.33 ± 0.67 b	0.67 ± 0.33 a	1.33 ± 0.67 b	2.00 ± 1.00 a	1.00 ± 0.58 b
T7 - Boveril® + Vessarya®	1.33 ± 0.33 a	0.00 ± 0.00 b	0.33 ± 0.33 a	2.67 ± 0.33 b	1.33 ± 0.88 a	2.00 ± 0.00 b
Second application (AA)[4]	-	-	-	-	-	-
T8 - Turbo® (AA)	0.67 ± 0.67 a	2.33 ± 1.33 b	0.67 ± 0.67 a	1.67 ± 0.67 b	1.67 ± 0.88 a	1.00 ± 0.58 b
T9 - Boveril® (AA)	1.33 ± 0.67 a	0.00 ± 0.00 b	1.67 ± 0.88 a	1.33 ± 0.33 b	1.00 ± 0.58 a	1.67 ± 0.67 b
T10 - Boveril® + Fox Xpro®	1.67 ± 1.20 a	2.33 ± 1.20 b	1.00 ± 0.00 a	0.00 ± 0.00 b	2.67 ± 2.19 a	0.67 ± 0.33 b
T11 - Boveril® + Sphere Max®	1.67 ± 0.33 a	0.67 ± 0.33 b	3.00 ± 0.58 a	2.67 ± 0.67 b	0.67 ± 0.33 a	2.00 ± 0.58 b
T12 - Boveril® + Orkestra®	1.00 ± 0.58 a	0.33 ± 0.33 b	0.67 ± 0.67 a	0.00 ± 0.00 b	2.67 ± 0.67 a	1.33 ± 0.67 b
T13 - Boveril® + Vessarya®	0.33 ± 0.33 a	1.00 ± 0.58 b	0.33 ± 0.33 a	1.00 ± 0.58 b	1.00 ± 0.00 a	1.00 ± 0.00 b
F (treatments)	0.99^{ns}	1.70*	0.99^{ns}	1.70*	0.99^{ns}	1.70*

Means followed by equal letters within a column do not differ statistically by the Scott-Knott test at 5% probability. Larval classification: Small larvae = neonate to 3rd instar, 1–10 mm; Large larvae = 4th to 5th instar, 11–25 mm; BA = applications at 9, 6, and 3 days before fungicide application; AA = applications at 3, 6, and 9 days after fungicide application (AA).

Table 5. Mean number (mean ± standard error) of live *Chrysodeixis includens* larvae observed in the 3rd evaluation, after application of the treatments in soybean, harvest (2024/2025), Chapadão do Sul, MS, Brazil.

Treatments	Small larvae	Large larvae	Small larvae	Large larvae	Small larvae	Large larvae
	9 days		6 days		3 days	
T1 - Control	4.33 ± 1.20 a	2.67 ± 0.67 a	4.33 ± 1.20 a	2.67 ± 0.67 a	4.33 ± 1.20 a	2.67 ± 0.67 a
First application (BA)3	-	-	-	-	-	-
T2 - Turbo®	3.67 ± 0.33 a	1.67 ± 0.88 a	2.67 ± 0.33 a	1.00 ± 1.00 a	2.00 ± 1.15 a	0.33 ± 0.33 a
T3 - Boveril®	1.00 ± 1.00 a	1.67 ± 0.33 a	5.33 ± 0.88 a	1.00 ± 0.58 a	3.00 ± 0.58 a	1.33 ± 0.88 a
T4 - Boveril® + Fox Xpro®	4.67 ± 1.86 a	3.33 ± 0.88 a	3.33 ± 0.67 a	0.67 ± 0.67 a	3.33 ± 0.88 a	2.00 ± 0.58 a
T5 - Boveril® + Sphere Max®	2.00 ± 1.00 a	3.00 ± 1.15 a	3.00 ± 0.58 a	1.00 ± 0.58 a	4.33 ± 2.33 a	3.00 ± 1.53 a
T6 - Boveril® + Orkestra®	2.67 ± 0.67 a	2.33 ± 0.88 a	3.00 ± 1.53 a	2.00 ± 0.58 a	2.00 ± 0.00 a	2.33 ± 1.20 a
T7 - Boveril® + Vessarya®	2.00 ± 0.58 a	0.33 ± 0.33 a	4.67 ± 0.88 a	1.00 ± 1.00 a	3.67 ± 1.86 a	1.00 ± 0.58 a
Second application (AA)4	-	-	-	-	-	-
T8 - Turbo®	3.00 ± 1.15 a	2.33 ± 0.88 a	1.00 ± 1.00 a	1.67 ± 0.88 a	1.33 ± 0.88 a	2.00 ± 0.58 a
T9 - Boveril®	1.67 ± 0.67 a	2.33 ± 0.88 a	3.33 ± 1.45 a	1.67 ± 0.33 a	2.00 ± 1.15 a	1.00 ± 0.58 a
T10 - Boveril® + Fox Xpro®	3.33 ± 0.33 a	2.67 ± 1.45 a	1.67 ± 1.20 a	1.33 ± 0.33 a	4.00 ± 2.08 a	1.00 ± 1.00 a
T11 - Boveril® + Sphere Max®	0.33 ± 0.33 a	3.33 ± 1.20 a	2.33 ± 1.86 a	1.67 ± 0.67 a	1.33 ± 0.33 a	1.67 ± 0.33 a
T12 - Boveril® + Orkestra®	1.67 ± 0.88 a	2.00 ± 0.58 a	3.67 ± 1.45 a	0.67 ± 0.33 a	2.00 ± 1.53 a	1.67 ± 0.33 a
T13 - Boveril® + Vessarya®	3.67 ± 1.76 a	1.33 ± 0.67 a	5.00 ± 2.52 a	2.00 ± 1.00 a	2.00 ± 0.58 a	1.33 ± 0.67 a
F. Treatments	1.00^{ns}	0.97^{ns}	1.00^{ns}	0.97^{ns}	1.00^{ns}	0.97^{ns}

Means followed by equal letters within a column do not differ statistically by the Scott-Knott test at 5% probability. Larval classification: Small larvae = neonate to 3rd instar, 1–10 mm; Large larvae = 4th to 5th instar, 11–25 mm; BA = applications at 9, 6, and 3 days before fungicide application; AA = applications at 3, 6, and 9 days after fungicide application (AA).

Table 6. Mean number (means ± standard error) of live *Chrysodeixis includens* larvae observed in the 4th evaluation, after application of the treatments in soybean crop (2024/2025), Chapadão do Sul, MS, Brazil.

Treatments	Small larvae	Large larvae	Small larvae	Large larvae	Small larvae	Large larvae
	9 days		6 days		3 days	
T1 - Control	4.67 ± 0.33a	2.33 ± 0.33 a	4.67 ± 0.33 A	2.33 ± 0.33 a	4.67 ± 0.33 a	2.33 ± 0.33 a
First application (BA)3						
T2 - Turbo®	2.00 ± 1.53a	1.00 ± 1.00 a	3.67 ± 0.88 a	0.00 ± 0.00 a	1.67 ± 0.33 a	0.33 ± 0.33 a
T3 - Boveril®	2.67 ± 1.20a	1.67 ± 0.67 a	2.33 ± 0.88 a	0.67 ± 0.33 a	1.00 ± 0.00 a	1.33 ± 0.33 a
T4 - Boveril® + Fox Xpro®	4.33 ± 1.86a	0.33 ± 0.33 a	3.00 ± 0.58 a	0.33 ± 0.33 a	2.33 ± 0.67 a	1.33 ± 0.33 a
T5 - Boveril® + Sphere Max®	1.00 ± 0.58a	0.00 ± 0.00 a	4.33 ± 1.20 a	1.00 ± 0.58 a	2.33 ± 0.67 a	1.67 ± 0.33 a
T6 - Boveril® + Orkestra®	4.67 ± 1.20a	1.33 ± 0.88 a	2.00 ± 0.58 a	0.67 ± 0.33 a	5.33 ± 1.45 a	2.00 ± 1.00 a
T7 - Boveril® + Vessarya®	4.33 ± 1.20a	1.00 ± 0.58 a	2.00 ± 2.00 a	0.67 ± 0.67 a	3.00 ± 0.58 a	1.33 ± 1.33 a
Second application (AA)4						
T8 - Turbo®	3.33 ± 0.67a	1.00 ± 1.00 a	3.00 ± 1.53 a	1.67 ± 0.33 a	2.00 ± 1.53 a	0.67 ± 0.33 a
T9 - Boveril®	3.67 ± 0.88a	1.33 ± 0.88 a	3.00 ± 1.15 a	1.33 ± 0.88 a	3.00 ± 2.08 a	1.33 ± 1.33 a
T10 - Boveril® + Fox Xpro®	3.67 ± 0.88a	2.00 ± 0.00 a	3.33 ± 1.45 a	1.33 ± 0.88 a	5.00 ± 1.53 a	1.33 ± 0.33 a
T11 - Boveril® + Sphere Max®	2.33 ± 0.88a	2.33 ± 0.33 a	3.00 ± 1.73 a	2.00 ± 0.58 a	1.33 ± 0.88 a	1.00 ± 0.58 a
T12 - Boveril® + Orkestra®	4.00 ± 1.53a	0.67 ± 0.67 a	2.33 ± 0.33 a	0.67 ± 0.33 a	4.33 ± 0.88 a	1.67 ± 0.88 a
T13 - Boveril® + Vessarya®	4.67 ± 0.67a	0.67 ± 0.33 a	2.00 ± 0.00 a	1.67 ± 1.67 a	4.00 ± 3.06 a	0.33 ± 0.33 a
F. Treatments	0.88^{ns}	0.79^{ns}	0.88^{ns}	0.79^{ns}	0.88^{ns}	0.79^{ns}

Means followed by equal letters within a column do not differ statistically by the Scott-Knott test at 5% probability. Larval classification: Small larvae = neonate to 3rd instar, 1–10 mm; Large larvae = 4th to 5th instar, 11–25 mm; BA = applications at 9, 6, and 3 days before fungicide application; AA = applications at 3, 6, and 9 days after fungicide application (AA).

Table 7. Mean numbers (means ± standard error) of live *Chrysodeixis includens* larvae observed in the 5th evaluation, after application of the treatments in soybean crop (2024/2025), Chapadão do Sul, MS, Brazil.

Treatments	Small larvae	Large larvae	Small larvae	Large larvae	Small larvae	Large larvae
	9 days		6 days		3 days	
T1 - Control	6.33 ± 1.45 a	2.00 ± 1.00 a	6.33 ± 1.45 a	2.00 ± 1.00 a	6.33 ± 1.45 a	2.00 ± 1.00 a
First application (BA)[3]						
T2 - Turbo®	3.00 ± 1.00 b	2.00 ± 0.58 a	3.67 ± 0.33 b	0.33 ± 0.33 a	3.00 ± 0.58 b	2.33 ± 1.20 a
T3 - Boveril®	7.67 ± 0.88 a	0.00 ± 0.00 a	10.00 ± 3.06 a	1.33 ± 0.88 a	3.00 ± 1.73 b	3.00 ± 1.53 a
T4 - Boveril® + Fox Xpro®	4.33 ± 1.76 b	1.67 ± 1.20 a	5.67 ± 1.45 a	1.33 ± 0.67 a	4.00 ± 1.15 b	1.00 ± 1.00 a
T5 - Boveril® + Sphere Max®	7.00 ± 1.15 a	1.67 ± 0.33 a	8.00 ± 2.08 a	1.00 ± 0.00 a	7.67 ± 2.73 a	0.33 ± 0.33 a
T6 - Boveril® + Orkestra®	6.67 ± 1.45 a	1.67 ± 0.88 a	4.67 ± 1.45 b	0.00 ± 0.00 a	6.33 ± 1.20 a	2.33 ± 0.33 a
T7 - Boveril® + Vessarya®	5.67 ± 0.33 a	0.67 ± 0.67 a	4.33 ± 2.33 b	2.00 ± 1.15 a	5.00 ± 0.58 b	0.33 ± 0.33 a
Second application (AA)[4]						
T8 - Turbo®	4.00 ± 1.15 b	1.00 ± 0.58 a	3.33 ± 0.67 b	0.33 ± 0.33 a	6.33 ± 1.20 a	1.33 ± 0.88 a
T9 - Boveril®	6.33 ± 0.33 a	1.67 ± 0.67 a	3.67 ± 0.67 b	1.00 ± 0.58 a	4.33 ± 1.33 b	0.33 ± 0.33 a
T10 - Boveril® + Fox Xpro®	7.67 ± 0.88 a	0.33 ± 0.33 a	3.33 ± 1.20 b	1.00 ± 1.00 a	5.33 ± 1.76 b	0.33 ± 0.33 a
T11 - Boveril® + Sphere Max®	1.33 ± 0.33 b	1.33 ± 0.88 a	5.33 ± 2.73 b	0.33 ± 0.33 a	4.33 ± 0.67 b	0.67 ± 0.33 a
T12 - Boveril® + Orkestra®	7.00 ± 1.73 a	1.00 ± 0.58 a	7.67 ± 0.88 a	2.33 ± 0.88 a	7.67 ± 2.73 a	0.67 ± 0.33 a
T13 - Boveril® + Vessarya®	3.33 ± 0.67 b	0.67 ± 0.33 a	7.00 ± 0.58 a	0.00 ± 0.00 a	4.67 ± 1.86 b	0.67 ± 0.67 a
F. Treatments	1.61*	1.22^{ns}	1.61*	1.22^{ns}	1.61*	1.22^{ns}

Means followed by equal letters within a column do not differ statistically by the Scott-Knott test at 5% probability. Larval classification: Small larvae = neonate to 3rd instar, 1–10 mm; Large larvae = 4th to 5th instar, 11–25 mm; BA = applications at 9, 6, and 3 days before fungicide application; AA = applications at 3, 6, and 9 days after fungicide application (AA).

Boveril® (positive control), Boveril® + Fox Xpro®, and Boveril® + Sphere Max® treatments showed significantly lower mean numbers of larvae than the other treatments (Table 7).

At the 3-day interval BA, the Turbo®, Boveril® (positive control), Boveril® + Fox Xpro®, and Boveril® + Vessarya® treatments showed significant reductions in small larvae. The Boveril® (positive control), Boveril® + Fox Xpro®, Boveril® + Sphere Max®, and Boveril® + Vessarya® treatments exhibited significantly lower means than the other treatments (Table 7).

4. Discussions

The untreated control exhibited higher mean numbers of both large and small larvae than most treatments across all evaluations and application intervals. However, the evaluated treatments (chemical insecticide, biological insecticide, and chemical fungicides) did not show a consistent pattern of interference with the efficacy of *B. bassiana*, as results varied across periods and larval sizes. During the first evaluation, no negative effects of fungicides applied in combination with Boveril® were identified, as all treatments showed variation in *C. includens* population density.

In the second evaluation, the mean number of large larvae in the untreated control differed significantly from that of the other treatments. The results of this evaluation indicate that combining Boveril® with fungicides reduced larval populations to levels similar to those of the positive controls (Boveril® alone; BA and AA). Larger larvae consume more leaf tissue and are, therefore, more exposed on the plant surface (Bueno et al., 2023; Padilha et al., 2025).

The lack of significant differences among treatments in the third and fourth evaluations may be related to the population reduction observed in the second evaluation. Entomopathogenic fungi can survive and disseminate conidia following parasitism of insects when temperature and humidity conditions are favorable (Mantzoukas et al., 2022; Souza et al., 2024; Martina et al., 2026).

The fourth evaluation preceded the 3- and 6-day interval applications of the pyrethroid insecticide and *B. bassiana*. These applications likely contributed to the observed reduction in pest population because pyrethroids exert a rapid contact effect that quickly decreases pest numbers (Ndakidemi et al., 2016; Rahman et al., 2024). In contrast, entomopathogenic fungi act more slowly, typically requiring approximately 3 days for colonization of the insect and 5 to 8 days for host death and sporulation (Loureiro et al., 2026).

In the fifth evaluation, no consistent pattern of fungicide interference with Boveril® efficacy was detected, and variation occurred in mean larval numbers depending on the interval (BA or AA). In this evaluation, higher numbers of small larvae, which are more susceptible than large larvae, were recorded. Balanced management during this period may have contributed to population reductions in subsequent pest generations.

Research on the compatibility of phytosanitary products with entomopathogenic fungi has produced variable results between in vitro and field conditions, because incompatibility observed in culture medium assays is not always replicated under field conditions (Loureiro et al., 2025). In studies evaluating the effects of pesticides on *B. bassiana*, incompatibility observed in culture medium assays between fungicides, insecticides, and *B. bassiana* was reduced by the soil microbiota when the products were applied together to the soil (Loureiro et al., 2002; Paiva-Guimarães et al., 2020).

Previous tests with the fungicides Fox Xpro®, Sphere Max®, Orkestra®, Vessarya®, and Elatus®, as well as the adjuvant Aureo®, demonstrated compatibility only of Elatus® and Aureo® with the fungus *M. rileyi* (Ascomycota: Clavicipitaceae), whereas Vessarya® completely inhibited colony growth (Loureiro et al., 2025). In the present study, the Vessarya® product applied in combination with Boveril® (BA and AA) resulted in population reductions similar to those observed in the other treatments. These results suggest that the fungicide did not interfere with the performance of the entomopathogen at the tested application intervals (Lacey et al., 2015; Zambolim, 2022; Faria et al., 2023).

The data from the present study showed that the products did not produce a consistent pattern of results, with variation across application intervals and soybean growth stages in relation to *C. includens* occurrence during the vegetative phase. Population reduction also varied depending on whether the products were applied before or after Boveril®.

The results of this study indicate that the evaluated fungicides did not exert negative effects on the pathogenic activity of *B. bassiana* against *C. includens* populations, because mean numbers of live larvae in all treatments and evaluation periods were not higher than those in the untreated control. Further studies are needed to expand knowledge on compatibility between entomopathogenic fungi and chemical products and to support the development of efficient and sustainable phytosanitary management strategies.

5. Conclusion

The fungicides tested at different application timings did not interfere with the efficacy of *B. bassiana* (Boveril®) in reducing populations of *C. includens* larvae.

Acknowledgements

The authors thank the National Council for Scientific and Technological Development (CNPq); the Federal University of Mato Grosso do Sul (UFMS), Campus Chapadão do Sul, MS, Brazil; the Federal University of Grande Dourados, Dourados, MS, Brazil; and the members of the LAMIP Research Group.

This study was partly financed by the Federal University of Mato Grosso do Sul (UFMS; Finance Code 001), the Brazilian Coordination for the Improvement of Higher Education Personnel (CAPES; Finance Code 001), and the Foundation to Support the Development of Education, Science and Technology of the State of Mato Grosso do Sul (FUNDECT-MS; Project process: 71/027.188/2022, no. 224/2022, SIAFEN: 31970, Process: 83/019.557/2025 no. 134/2025 SIAFIC: 2638; and the granting of the master's scholarship, Process: 83/003.581/2024, no. 90/2024).

Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

References

BHERING, L.L., 2017. Rbio: a tool for biometric and statistical analysis using the R platform. *Crop Breeding and Applied Biotechnology*, vol. 17, no. 2, pp. 187-190. https://doi.org/10.1590/1984-70332017v17n2s29.

BORGES, F.S.P., LOUREIRO, E.S., JAURRETCHE, J.E., PESSOA, L.G.A., ARRUDA, L.A., DIAS, P.M. and NAVARRETE, A.A., 2021. Performance of phytosanitary products for control of soybean caterpillar. *Anais da Academia Brasileira de Ciências*, vol. 93, no. 4, pp. e20200205. https://doi.org/10.1590/0001-3765202120200205. PMid:34705937.

BUENO, A.F., COLMENAREZ, Y.C., CARNEVALLI, R.A. and SUTIL, W.P., 2023. Benefits and perspectives of adopting soybean-IPM: the success of a Brazilian programme. *Plant Health Cases*, vol. 2023, pp. 1-16.

EMBRAPA. (2000). *Embrapa Soybean Technical recommendations for soybean cultivation in the central region of Brazil*. Londrina: Embrapa Soja/MT Foundation, 245 pp. (Embrapa Soybean, n. 146).

FARIA, M., MASCARIN, G.M., BUTT, T. and LOPES, R.B., 2023. On-farm production of microbial entomopathogens for use in agriculture: Brazil as a case study. *Neotropical Entomology*, vol. 52, no. 2, pp. 122-133. https://doi.org/10.1007/s13744-023-01033-5. PMid:37014592.

FILGUEIRAS, M.D.G., MASCARIN, G.M., LIMA, V.H., BARRETO, L.P., DE SOUZA RIBEIRO-SILVA, C.S., PINTO, S.M.N., RIZZO, P.V., FREITAS, F.M.C., SOARES, F.E.F., MONTEIRO, C. and FERNANDES, E.K.K., 2025. Harnessing environmental microbiological interventions with micro- and macroorganisms for assessing cattle tick management. *Scientific Reports*, vol. 15, no. 1, pp. 15836. https://doi.org/10.1038/s41598-025-90123-y. PMid:40328925.

GONÇALVES, J., CALIXTO, E.S., DE FREITAS BUENO, A., DOURADO, P.M. and PAULA-MORAES, S.V., 2024. Oviposition patterns of primary lepidopteran defoliators in soybean and the impact on structured refuge recommendations. *Pest Management Science*, vol. 80, no. 11, pp. 5619-5629. https://doi.org/10.1002/ps.8277. PMid:38940546.

LACEY, L.A., GRZYWACZ, D., SHAPIRO-ILAN, D.I., FRUTOS, R., BROWNBRIDGE, M. and GOETTEL, M.S., 2015. Insect pathogens as biological control agents: back to the future. *Journal of Invertebrate Pathology*, vol. 132, pp. 1-41. https://doi.org/10.1016/j.jip.2015.07.009. PMid:26225455.

LOUREIRO, E.S., BARBOSA JÚNIOR, G.B., DIAS, P.M., PESSOA, L.G.A., RODRIGUES, A.S., GODOY, M.S., MESA, N.E.S. and GREGORI, G.S., 2025. In vitro compatibility of fungicides with *Metarhizium rileyi* in soybean disease management. *Revista Caatinga*, vol. 38, pp. e12752. https://doi.org/10.1590/1983-21252025v3812752rc.

LOUREIRO, E.S., DIAS, P.M., PESSOA, L.G., AMARAL, T.S.D., PESSOA, M.B., GREGORI, G.S., RODRIGUES, A.S. and GODOY, M.S., 2024.

Virulence of entomopathogenic fungi in larvae of Lepidoptera: noctuidae. *Revista Caatinga*, vol. 37, pp. 1-7. https://doi.org/10.1590/1983-21252024v3712375rc.

LOUREIRO, E.S., MOINO, J.R.A., ARNOSTI, A. and SOUZA, G.C., 2002. Effect of chemical phytosanitary products used in lettuce and chrysanthemum on entomopathogenic fungi. *Neotropical Entomology*, vol. 31, no. 2, pp. 263-269. https://doi.org/10.1590/S1519-566X2002000200014.

LOUREIRO, E.S., OLIVEIRA, G.S., RODRIGUES, A.S., PESSOA, L.G.A., AMARAL, T.S., PESSOA, M.B., GREGORI, G.S., SOTO-MESA, N.E. and DIAS, P.M., 2026. Potential of adjuvants and an entomopathogenic fungus for the management of *Helicoverpa armigera*. *Brazilian Journal of Biology*, vol. 86, pp. e303128. PMid:41849476.

MANTZOUKAS, S., KITSIOU, F., NATSIOPOULOS, D. and ELIOPOULOS, P.A., 2022. Entomopathogenic fungi: interactions and applications. *Encyclopedia*, vol. 2, no. 2, pp. 646-656. https://doi.org/10.3390/encyclopedia2020044.

MAPA/AGROFIT, 2025 [viewed 23 March 2026]. *Phytosanitary Pesticide Systems - Open Consultation* [online]. Available from: http://agrofit.agricultura.gov.br/agrofit_cons/principal_agrofit_cons.

MARTINA, T.N., VEYB, R.T., STECCAA, J.D.L., BRUMA, L.N., CIPRIANIA, L.P. and BRASIL NETO, E.S., 2026. Applications of *Trichoderma asperelloides* in soybean production: a perspective based on bibliometric and experimental data. *Brazilian Journal of Biology*, vol. 86, pp. e301828. https://doi.org/10.1590/1519-6984.301828. PMid:41880388.

MASCARIN, G.M., SHRESTHA, S., DUNLAP, C.A., RAMIREZ, J.L. and COLEMAN, J.J., 2025. Enhanced virulence through genetic engineering of *Beauveria bassiana* blastospores by overexpression of a cuticle-degrading endochitinase. *Journal of Invertebrate Pathology*, vol. 213, pp. 108420. https://doi.org/10.1016/j.jip.2025.108420. PMid:40780391.

NDAKIDEMI, B., MTEI, K. and NDAKIDEMI, P.A., 2016. Impacts of synthetic and botanical pesticides on beneficial insects. *Agricultural Sciences*, vol. 07, no. 6, pp. 364-372. https://doi.org/10.4236/as.2016.76038.

OLIVEIRA, C.M., FARIA, M.R. and LOPES, R.B., 2025. Does *Beauveria bassiana* infection in *Dalbulus maidis* reduce the transmission of phytoplasma and spiroplasma and the severity of corn stunt diseases? *Crop Protection (Guildford, Surrey)*, vol. 197, pp. 107361. https://doi.org/10.1016/j.cropro.2025.107361.

PADILHA, K.S.M.A., PEGORARO, R.F., BARROSO, A.M.F., SOARES, V.A.S.F., ALVES, P.G.L., FERREIRA, E.A. and SOUZA, R.P.D., 2025. Damage caused by caterpillars in chickpea crops in tropical regions. *Brazilian Journal of Biology*, vol. 85, pp. e290139. https://doi.org/10.1590/1519-6984.290139. PMid:40435073.

PAIVA-GUIMARÃES, A.G.L., FREIRE, K.R.L., SANTOS, S.F.M., ALMEIDA, A.F. and SOUSA, A.C.B., 2020. Alternative substrates for conidiogenesis of the entomopathogenic fungus *Beauveria bassiana* (Bals) Vuillemin (Deuteromycotina: hyphomycetes). *Brazilian Journal of Biology*, vol. 80, no. 1, pp. 133-141. https://doi.org/10.1590/1519-6984.195711. PMid:31017241.

RAHMAN, M.D.M., OMOTO, C. and KIM, J., 2024. Genome-wide exploration of long non-coding RNAs of *Helicoverpa armigera* in response to pyrethroid insecticide resistance. *Insects*, vol. 15, no. 3, pp. 146. https://doi.org/10.3390/insects15030146. PMid:38535342.

SENI, A., 2023. Potential of the various oils for insect pests' management and their effect on beneficial insects. *International Journal of Tropical Insect Science*, vol. 43, no. 2, pp. 321-337. https://doi.org/10.1007/s42690-023-00970-3.

SOUZA, T.D., GODOY, S.M., FELICIANO, D.C., BINNECK, E., RANGEL, D.E.N. and SOSA-GÓMEZ, D.R., 2024. Genetic diversity of the entomopathogenic fungus *Metarhizium rileyi* based on de novo microsatellite markers. *Journal of Invertebrate Pathology*, vol. 204, pp. 108081-10. https://doi.org/10.1016/j.jip.2024.108081. PMid:38458349.

VELOZO, S.G.M., CARVALHO, V.R., TEIXEIRA, D.L., PASSOS, J.R.S., ZANUNCIO, J.C., STEPHAND, D. and WILCKEN, C.F., 2025. Effect of temperature and substrate on conidia yield of entomopathogenic fungi using the JKI solid-state bioreactor system. *Brazilian Journal of Biology*, vol. 85, pp. e295209. https://doi.org/10.1590/1519-6984.295209. PMid:41337534.

WANG, Y., YANG, W.C., CLERCQ, P., LEPPLA, N.C., BUENO, A.F., RAMIREZ-ROMERO, R., DESNEUX, N. and ZANG, L.S., 2026. Biotechnology-driven artificial diets for mass-rearing arthropod natural enemies. *Trends in Biotechnology*, vol. 20, pp. 1-15. https://doi.org/10.1016/j.tibtech.2025.12.010. PMid:41577521.

ZAMBOLIM, L., 2022. Integrated management of Asian soybean rust. *European Journal of Applied Sciences*, vol. 10, no. 2, pp. 602-633. https://doi.org/10.14738/aivp.102.12215.