




# Dual Impact Assessments of *Beauveria bassiana* (Ascomycota: Hypocreales) Against *Brevicoryne brassicae* (Hemiptera: Aphididae)

Patient Niyibizi Gakuru<sup>1,2,3</sup>  · Marcellin Cuma Cokola<sup>1,4</sup> · Kenza Dessauvages<sup>1</sup> · Jean-Pierre Mate Mweru<sup>2</sup> · François Muhashy Habiyaremye<sup>3</sup> · Rudy Caparros Megido<sup>1</sup> · Frédéric Francis<sup>1</sup> · Ibtissem Ben Fekih<sup>1</sup>

Received: 16 June 2025 / Accepted: 20 September 2025  
© Sociedade Entomológica do Brasil 2025

## Abstract

Entomopathogenic fungi (EPF) are promising eco-friendly alternatives to chemical insecticides against various insect pests. We assessed the efficacy of different *Beauveria bassiana* (Bals.-Criv.) Vuill. strains against *Brevicoryne brassicae* (L.), a major pest of Brassica crops under both laboratory and field conditions. The virulence and endophytic properties of three strains of *B. bassiana*, the commercial strain GHA, the endemic one KA14 from the Democratic Republic of Congo (DRC), and GxABT-1 from Belgium, were assessed under laboratory conditions. Under field conditions in eastern DRC, the efficacy of the *B. bassiana* GHA was evaluated compared to that of a conventional insecticide. In vitro, the lethal time 50 (LT<sub>50</sub>) values ranged from 3 days for *B. bassiana* GxABT-1 to 4 days for both GHA and KA14 following direct application of fungi. Aphid exposure to fungus-treated leaves at 7 and 14 days after inoculation (DAI) resulted in LT<sub>50</sub> values of 4 to 5 days for GHA and KA14. Regarding the endophytic potential, *B. bassiana* KA14 showed higher performance in colonizing plant tissues and improving plant height growth. In the dual-choice tests, aphids were initially attracted to fungus-treated plants at 7 DAI but showed no preference between treated and control plants at 14 DAI. Field trials indicated no significant difference between fungal and chemical insecticide treatments, but both significantly reduced *B. brassicae* infestation compared to the Control. Our results support the interest of exploring the potential of the endemic *B. bassiana* KA14 strain against various insect pests and applying different methods.

**Keywords** Cabbage aphid · Entomopathogenic fungi · Hypocreales · Endophyte · Microbial control

---

Edited by María F Cingolani

✉ Patient Niyibizi Gakuru  
patientgakuru@gmail.com

- <sup>1</sup> Functional and Evolutionary Entomology, TERRA, Gembloux Agro-Bio Tech, Univ of Liège, Gembloux, Belgium
- <sup>2</sup> Regional Post-Graduate Training School On Integrated Management of Tropical Forests and Lands (ERAIFT), POBox 15373, Kinshasa, Democratic Republic of Congo
- <sup>3</sup> Dept of Crop Sciences, Faculty of Agriculture and Environmental Sciences, Univ of Goma, POBox 204, Goma, Democratic Republic of Congo
- <sup>4</sup> Dept of Crop Sciences, Faculty of Agriculture and Environmental Sciences, Univ Evangelique en Afrique, POBox 3323, Bukavu, Democratic Republic of Congo

## Introduction

The cabbage aphid, *Brevicoryne brassicae* (Linnaeus, 1758) (Hemiptera: Aphididae), is an important limiting pest causing severe losses in Brassica crops (Pereira et al. 2019; Mpumi et al. 2020; Gakuru et al. 2024). The highly damaging potential of *B. brassicae* is mainly due to its reproductive capacity, especially in warm weather (Munthali and Tshego-fatso 2014), and its feeding activity, which causes the wilting of leaves and therefore a poor head formation (Saha et al. 2018; Hao et al. 2019).

Chemical insecticides are the main practice used by vegetable growers to protect cabbage from insects (Tudi et al. 2021). In the southeastern Democratic Republic of Congo (DRC), 86% of smallholder farmers in Lubumbashi rely on chemical control practices to protect the Chinese cabbage crops (Balasha and Nsele 2019). Such practices cause adverse effects on the environment and human health. They can also be toxic to non-target organisms, including beneficial insects, besides

the emergence of resistance within treated insect pests (Ntow et al. 2006; Tudi et al. 2021). As an alternative control method, biological control agents are being encouraged in Integrated Pest Management (IPM) (Vu et al. 2007; Prabha et al. 2016). Beneficial microbes such as fungi are among the sustainable and eco-friendly alternatives to chemical pesticides within IPM systems, enhancing crop protection while minimizing environmental impact. Due to their specificity and ability to infect through the cuticle, hypocrealean entomopathogenic fungi (EPF) exhibit a broad host range of arthropods, including sap-sucking insects such as aphids (Biryol et al. 2022; Shang et al. 2024). For example, *Beauveria bassiana* (Bals.-Criv.) Vuill. (Hypocreales: Cordycipitaceae) is one of the most studied species within EPF. It exhibits a cosmopolitan distribution and affects many insect hosts (Mascarin and Jaronski 2016). Besides its entomopathogenic potential, *B. bassiana* is also recognized as an endophytic fungus that can colonize plant tissues without harming its host (Francis et al. 2022). In this form, a mutualistic relationship is established in which the fungus benefits from plant-derived nutrients and, in response, promotes plant growth and enhances the host immune system against biotic stressors, including pathogens and insect pests (Torkaman et al. 2023).

Many strains of *B. bassiana* have been used to control vegetable insect pests, including the diamondback moth *Plutella xylostella* (Linnaeus, 1758) (Shehzad et al. 2022), the cabbage looper *Trichoplusia ni* (Hübner, 1803) (Mireles-Valdez et al. 2023), the green peach aphid *Myzus persicae* (Sulzer, 1776) (Yogananda et al. 2023; Arinanto et al. 2024), and the tomato fruit worm *Helicoverpa armigera* (Hübner, 1808) (Qayyum et al. 2015). However, little attention has been paid to their performance against *B. brassicae*. Different studies have evaluated the biocontrol properties of the *B. bassiana* GHA strain in different agroclimatic areas around the world (Świergiel et al. 2016; Mantzoukas et al. 2022), but to our knowledge, no study has been conducted to evaluate their potential against the cabbage aphid under the environmental conditions of eastern DRC.

This study aims to evaluate the virulence of different strains of *B. bassiana* against *B. brassicae*. Specifically, the study investigated (a) the pathogenicity of different *B. bassiana* strains on *B. brassicae* as well as their endophytic potential under laboratory conditions and (b) the performance of the commercial *B. bassiana* GHA against the same aphid species under natural conditions of eastern DRC. The outcome of such a study will help to establish more sustainable practices to control *B. brassicae*.

## Materials and methods

### Evaluation of the pathogenicity and endophytic behavior of entomopathogenic fungi under laboratory conditions

#### Cabbage plants and aphids rearing

Healthy cabbage plants were obtained by direct seeding onto seed trays filled with a universal potting soil (TERS50, La Plaine Chassart, Belgium). Two weeks after sowing, the seedlings were transplanted into 10-cm diameter pots. Both trays and pots were placed in closed cages in a growth chamber under controlled conditions at  $23 \pm 1$  °C, 60% RH, and LD 16:8 h. Colonies of *B. brassicae* were collected from infested cabbage crops and transferred to the Functional and Evolutionary Entomology Laboratory at Gembloux Agro-Bio Tech (University of Liège, Belgium). Healthy aphids were then picked up from the samples and used to infest six-leaf stage healthy cabbage plants and kept in rearing cages (BugDorm 4M4545, 90 MegaView Science Co. Ltd., Taichung, Taiwan) under the same controlled conditions as mentioned above. Several successive generations were monitored to ensure the aphid colonies were free of parasitoids or natural fungal infection.

#### *Beauveria bassiana*: origin and inoculum preparation

A total of three strains of *B. bassiana* were used in this study, including the GHA strain isolated from the commercial formulation Botanigard 22WP, GxABT-1 isolate obtained from soil samples as described in Dessauvages et al. (2024), while KA14 is an endemic strain isolated from earwig (Dermaptera: Forficulidae) cadavers collected in eastern DRC (Cokola et al. 2023). The different *B. bassiana* strains were maintained on Sabouraud Dextrose Agar (SDA) media and incubated for 2 weeks at  $23 \pm 1$  °C to allow mycelia and spore production. The fungal inoculum was prepared by harvesting the different fungal structures (conidia and mycelia) into 10 mL of 0.03% Tween 80 using a sterilized spatula. The fungal suspension was then filtered using sterile cheesecloth. Conidia count was determined using a Bürker hemocytometer (Marienfeld, Germany), and concentration was adjusted to  $1 \times 10^8$  mL<sup>-1</sup>. To test the viability of the conidia of each strain, 100 µL of  $1 \times 10^5$  conidia mL<sup>-1</sup> dilution was spread on an SDA plate and incubated at  $23 \pm 1$  °C. After 24 h, germinated and non-germinated conidia were counted, and only when we reported > 90% viability were used for the bioassays.

### Efficacy of fungal treatments against *Brevicoryne brassicae*

Ten aphids were placed in a Petri dish and sprayed, from a 10 cm distance, with 1 mL of a  $1 \times 10^8$  conidia  $\text{mL}^{-1}$  of fungal inoculum (*B. bassiana* GHA, KA14 and GxABT-1). The same number of aphids was sprayed with 0.03% Tween 80 for the negative control. Each group of treated aphids was then transferred to a young cabbage leaf taken from non-infested 3-week-old seedlings secured in a plastic cup containing 1.5% agar solution, following the method by Ben Fekih et al. (2019). The agar served as a micro-humid environment to keep the aphid's food resource fresh during the bioassay. Five replicates were performed for each treatment. The different cups were kept in controlled conditions at 23 °C for LD 16:8 h. Aphid mortality was checked daily over 8 days. Aphid cadavers were collected daily and prepared for surface sterilization by immersion in 70% ethanol for 10 s and then washed three times for 10 s in sterile water, as described in Dessauvages et al. (2024). Then, the cadavers were placed on sterile wet filter paper in a Petri dish and incubated at 23 °C to allow fungal outgrowth if it existed.

### Endophytic potential and indirect impact on *Brevicoryne brassicae*

Both *B. bassiana* GHA and the endemic strain KA14 conidia suspensions were prepared as described above. Five milliliters of  $1 \times 10^8$  conidia  $\text{mL}^{-1}$  of each of the fungal suspensions was used to spray the leaves and stem of 3-week-old cabbage plants following the method of Jaber and Enkerli (2017) and Jaber et al. (2018). The control plants were sprayed with an equal volume of 0.03% Tween 80. Twenty plants were used to assess the indirect impact of the *B. bassiana* treatments against the aphid. The endophytic impact of *B. bassiana* GHA and KA14 in treated plants on *B. brassicae* was tested at 7 and 14 days after inoculation (DAI). Cabbage leaves harvested at 7 and 14 DAI were individually secured in 1.5% agar solution in cups of 30 mL. Then, 10 aphids were transferred onto each of the leaves. The mortality of aphids was checked daily for a period of 8 days, as above. Five replicates were adopted for each treatment and sampling time.

### Impact of fungal treatments on plant growth and endophytic colonization

Growth parameters such as plant height (the distance from the base of the plant to the tip of the longest leaf) were measured at 7 and 14 DAI for both inoculated and control plants. The experiment followed a three-treatment design (two strains of *B. bassiana* GHA and KA14 and the Control); each treatment included a total of 20 plants, 10 of which were sampled at 7 DAI and then at 14 DAI for plant growth

parameters assessment and endophytic plant colonization assays. The net height increase parameter, adapted from Torkaman et al. (2023), was estimated as the difference between the height at 7 or 14 DAI and the initial height before inoculation. The endophytic potential of both strains of *B. bassiana* on treated plants was assessed at 7 and 14 DAI following Parsa et al. (2013). The same plants chosen randomly for assessing the growth parameters were used to perform the endophytic colonization assessment. Leaves, stems, and roots from five plants per treatment were processed on each sampling day. Plant material was surface sterilized by immersion in 70% ethanol for 2 min, followed by 2 min in sodium hypochlorite and three rinses in sterile distilled water. The plant material was surface-dried in a laminar flow hood. Six leaf discs (approximately 1  $\text{cm}^2$ ) were cut from surface-sterilized leaves, and a similar number of stem tissue pieces (approximately 1 cm long) were obtained using a sterile scalpel. Six selected segments (approximately 1 cm long) of the different root parts (primary roots and lateral roots) were cut using a sterile scalpel. The six sampled plant tissues from each plant segment were then evenly distributed on separate Petri dishes containing SDA culture medium supplemented with streptomycin (0.5 mL of 0.6  $\text{g mL}^{-1}$ ), tetracycline (0.5 mL of 0.05  $\text{g mL}^{-1}$ ), and cyclohexamide (1 mL of 0.05  $\text{g mL}^{-1}$ ). The dishes were incubated at  $25 \pm 1$  °C and inspected for fungal outgrowth after 14 days. The percent colonization frequency (CF) was calculated using the following formula from Altaf et al. (2023).

$$\text{CF} = \frac{\text{Number of plant segments showing fungal growth}}{\text{Total number of plated plant segments}} \times 100$$

### Dual-choice tests

To assess the impact of endophytic colonization of the tested fungal strains on aphid behavior, leaf discs from cabbage plants were used for dual-choice assays, as described in Fingu-Mabola et al. (2020). Leaf discs of 1 cm diameter were collected from inoculated cabbage plants at 7 and 14 DAI and control plants. The different leaf discs were taken from leaves of the same age per plant at both sampling times and placed in 9-cm diameter Petri dishes containing moist filter paper (VWR Grade 424, 90 mm diameter, Germany). The Petri dishes were kept in the dark for 10 min to reduce the emission of volatiles from the cutting procedure as explained in Fingu-Mabola et al. (2020). Then, two leaf discs of two combinations (fungal treated at 7 DAI or 14 DAI and Control) were placed about 6 cm apart. A total of 10 adult aphids kept for 2 h without food were placed in the center of the Petri dish to assess feeding preference. The dishes were kept in a ventilated incubator at

23°C. The number of aphids on each of the two discs was recorded after 2, 4, and 6 h. Each combination of choice was repeated five times.

## Field experiments

Field experiments were conducted in eastern DRC, North-Kivu province, Nyiragongo Territory, about 15 km north of Goma city during two consecutive seasons: an experiment conducted in the long rainy season (October 2022–January 2023) named “LRS” and an experiment conducted in the short rainy season (March–July 2023) named “SRS.” Geographic coordinates are 1°36.4544'S; 29°15.4049'E; altitude 1801 m and 01°36.3277'S; 29°15.4736'E; altitude 1815 m, respectively. For each experimental season, a completely randomized block design was used with three replicates for each treatment: a commercial formulation of *B. bassiana* GHA (Botanigard 22WP), the pyrethroid chemical insecticide (Cyperscope 5EC) as a positive control, and the negative control plots treated only with water (Table 1). The wettable powder of Botanigard 22WP was applied according to EU standards (Certis 2021). The first insecticide application was made at the vulnerable stage of the crop to insect attacks, i.e., 15 days after transplanting (DAT). Two further applications were made at about 10-day intervals, depending on weather conditions, particularly rainfall. All insecticide applications were made with a backpack sprayer. The plots selected for each treatment were 4 × 2 m and contained 24 plants each. Transplanting was carried out on raised beds with a 0.5 m × 0.4 m spacing. No agrochemical product was applied, but all plots received an organic fertilizer. Other recommended cultural operations, such as weeding and ridging, were applied evenly using hand tools. Cabbage plants within plots were monitored every 7 days from 7 DAT, and aphids were counted using a 10× magnifying glass. Spaces between leaves and their upper and lower surfaces were also checked for hidden insects (Shonga and Getu 2021; Mantzoukas et al. 2022). General plant health was assessed by counting the number of plants showing signs of aphids. The following formula was used to determine the infestation rate:

$$\text{Infestation rate} = \frac{\text{Number of infested plant}}{\text{Total number of plants}} \times 100$$

## Statistical analysis

Statistical analyses were performed using R 4.4.1 software. First, a descriptive analysis was carried out to identify general patterns in the data. Using the Kaplan-Meier method, survival probabilities at each event time were estimated using the *survival* and *survminer* packages (Kassambara et al. 2016; R Core Team 2024; Therneau 2024), considering the number of aphids exposed to entomopathogenic fungi and the number of observed events to generate the survival curve. The log-rank test was then used to assess differences in survival between treatments. The effect of entomopathogenic fungal treatments on endophytism rate was assessed using the Kruskal-Wallis test. When a significant difference between treatments was observed, post hoc comparisons were made using Dunn's test with Bonferroni correction. Medians, quartiles, and interquartiles of the endophytic rate were calculated, and box plots were used to visualize the distributions. The endophytic potential was analyzed using a beta regression generalized linear model (GLM) with a logit link function, with segment, treatment, and day after inoculation as explanatory variables. Since the endophytic rate is expressed as a percentage (bounded between 0 and 100), a transformation was made using the Smithson-Verkuilen adjustment (Smithson and Verkuilen 2006) to meet the requirements of the model. Analysis of variance (ANOVA) was performed to test the overall effect of each explanatory variable on the response variable. For significant main effects, pairwise comparisons were conducted using estimated marginal means with Tukey's honest significant difference (HSD) adjustment for multiple testing. A similar ANOVA was applied to assess the effect of treatment on plant height. A generalized linear mixed model (GLMM) was applied for the choice test using the *glmer* function. The model assessed the effects of treatment and time on the response variable (number of insects per treatment), assuming a Poisson distribution with a log-link function. In the field experiments, a two-way ANOVA was performed to evaluate the effect of treatment and time on aphid infestations at 5% significance. The interaction between treatment and time was also tested. When a significant difference was observed, post hoc comparisons were made using Tukey's test. The significance level was set at 5% for all tests. All graphs were generated using the R package *ggplot2* (Wickham 2016).

**Table 1** Description of insecticides used in the field experiments

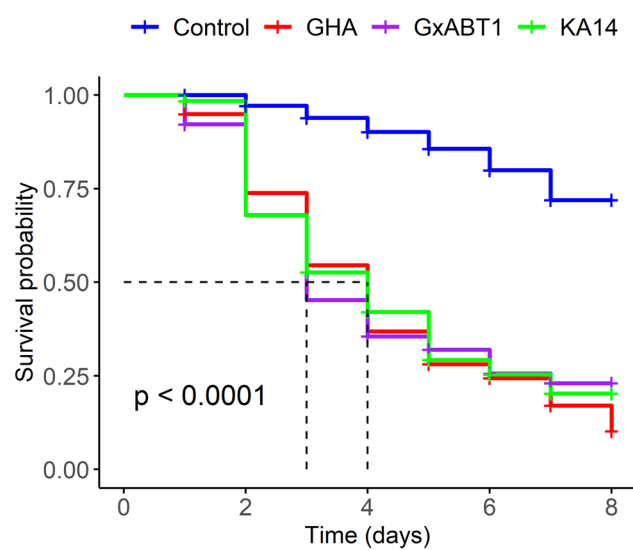
Commercial name	Description/active ingredient	Dose
Botanigard 22WP	Entomopathogenic fungus, <i>Beauveria bassiana</i> , GHA strain; $4.4 \times 10^{10}$ viable conidia/kg	0.0625%, i.e., 62.5 g/100 L of water
Cyperscope 5EC	Pyrethroid insecticide, Cypermethrin 5%	100 mL/100 L of water
Control	Water	-

## Results

### Lethal effect and endophytic activity of *Beauveria bassiana* strains under laboratory conditions

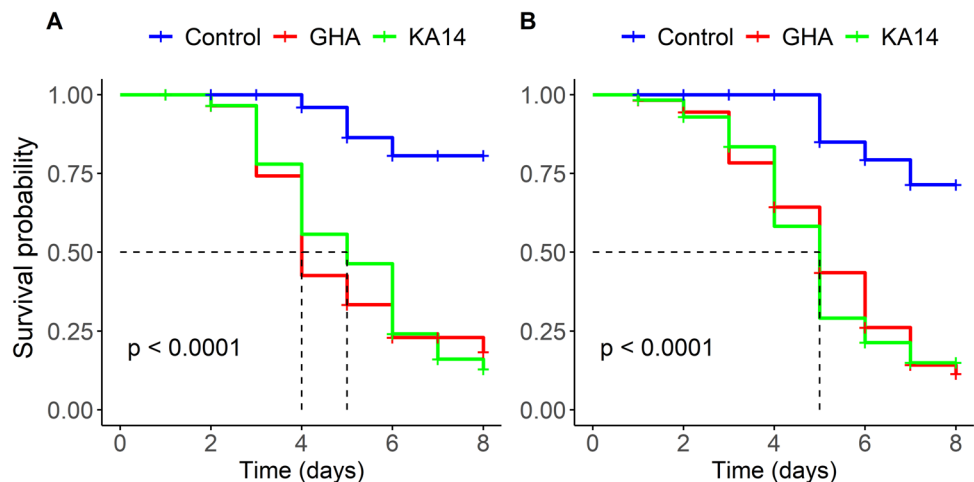
#### Effect of fungal treatments on *Brevicoryne brassicae*

The Kaplan-Meier survival analysis revealed significant differences in survival probability among all groups following direct application of entomopathogenic fungi ( $p < 0.001$ ; 95% confidence level), as shown in Fig. 1. All treated groups (GxABT-1, GHA, and KA14) showed a steep decline in survival, with the lethal time 50 (LT<sub>50</sub>) values of 3 days for



**Fig. 1** Kaplan-Meier survival probability following direct application of *Beauveria bassiana* to adult *Brevicoryne Brassicae*. The log-rank test was used for significant differences between treatments ( $p < 0.05$ ). Survival rates were calculated based on five replicates for each treatment

**Fig. 2** Kaplan-Meier survival probability of adult *Brevicoryne brassicae* exposed to *Beauveria bassiana*. **A** Survival probability following insect exposure to fungus-treated leaves harvested 7 days after inoculation; **B** Survival probability following insect exposure to fungus-treated leaves harvested 14 days after inoculation. The log-rank test was used for significant differences between treatments ( $p < 0.05$ ). Survival rates were calculated based on five replicates for each treatment



GxABT-1 and 4 days for both GHA and KA14. In contrast, the control group showed a much slower decline, with over 75% survival maintained beyond 4 days, confirming that mortality was treatment-induced.

#### Indirect effects of fungal treatments on *Brevicoryne brassicae*

The Kaplan-Meier survival analysis revealed significant differences in survival probability following exposure to fungus-treated leaves collected at 7 and 14 DAI (Fig. 2). Both GHA and KA14 treatments caused rapid mortality, with median survival time dropping sharply within 4 and 5 days of exposure to treated leaves collected at 7 and 14 DAI, respectively. In contrast, the control group maintained high survival (>70% throughout the observation period). The curves for GHA and KA14 overlap closely, suggesting no significant difference in their efficacy.

#### Effects of fungal treatments on cabbage plant growth parameters

The effects of the endophytic fungal treatments and the time after inoculation on the net height increase (NHI) are shown in Table 2. No significant differences between treatments were observed for the NHI recorded 7 days after inoculation (DAI). The treatments had no measurable effect on plant height under the conditions tested ( $F_{(2,12)} = 0.12$ ,  $p = 0.886$ ). However, a significant difference exists between treatments in NHI calculated at 14 DAI. Seedlings treated with endophytic fungi solutions show higher values than controls ( $F_{(2,12)} = 5.73$ ,  $p = 0.017$ ).

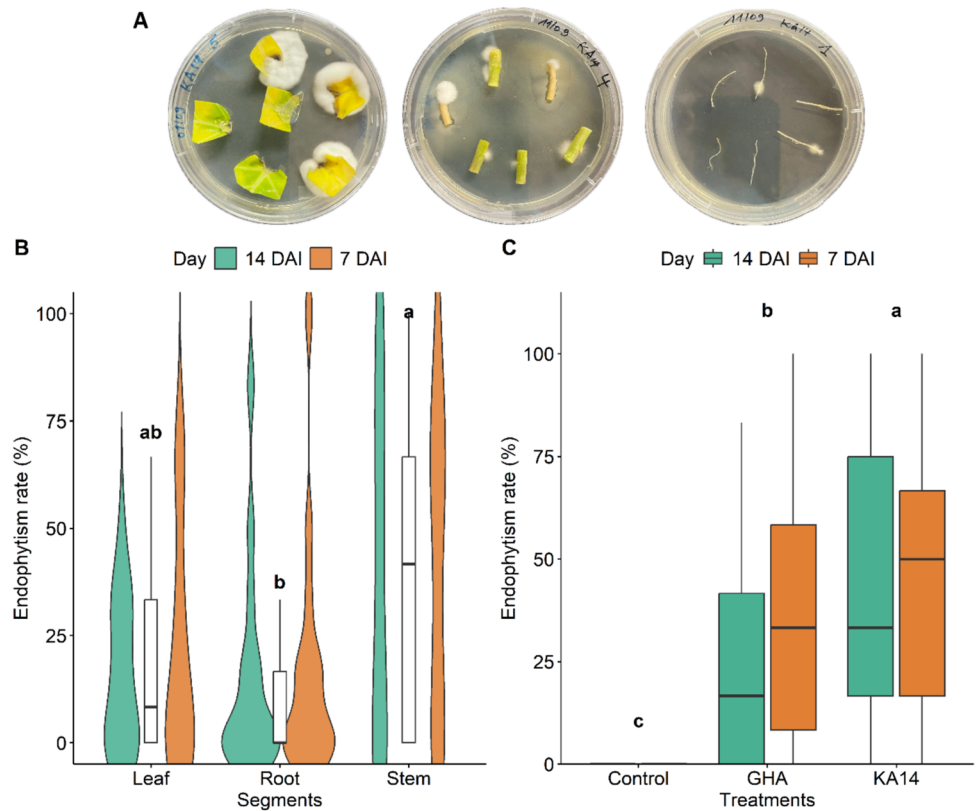
The potential of the different fungal strains to colonize the different plant parts was registered at 7 and 14 DAI (Fig. 3). A Kruskal-Wallis test indicated significant differences in colonization across treatments ( $H_{(2)} = 43.3$ ,  $p < 0.001$ ) and among plant segments ( $H_{(2)} = 6.7$ ,  $p = 0.033$ ). Beta

**Table 2** Effect of endophytic *Beauveria bassiana* isolates on the plant height

Days after inoculation	Treatment	Plant height before inoculation (cm)	Plant height after inoculation (cm)	Net height increase	<i>p</i> -value
7 DAI	<i>B. bassiana</i> GHA	17.6 ± 1.51	19.2 ± 1.68	1.6 ± 0.22 <sup>a</sup>	0.886
	<i>B. bassiana</i> KA14	17.6 ± 1.38	19.1 ± 2.24	1.5 ± 0.93 <sup>a</sup>	
	Control	17.7 ± 1.39	19.1 ± 1.71	1.4 ± 0.54 <sup>a</sup>	
14 DAI	<i>B. bassiana</i> GHA	18.4 ± 1.08	21.6 ± 1.24	3.2 ± 0.27 <sup>a</sup>	0.017*
	<i>B. bassiana</i> KA14	16.9 ± 2.30	20.3 ± 2.41	3.4 ± 0.83 <sup>a</sup>	
	Control	16.8 ± 0.90	18.8 ± 1.03	2.0 ± 0.50 <sup>b</sup>	

Means ± standard deviation followed by the same letter do not differ statistically at 5% significance according to the Tukey post hoc test, and an asterisk shows significant differences between treatments: \**p* < 0.05

**Fig. 3** Endophytic potential of *Beauveria bassiana* isolates in cabbage plant. **A** *Beauveria bassiana* growing from cabbage tissues. From left to right, leaf, stem, and root segments are shown. **B** Violin plot representing the endophytism rate of fungal isolates from plant segments collected at 7 and 14 days after inoculation (DAI). **C** Box plot of endophytism rate according to treatments at 7 and 14 DAI. The significance letters (a, b) indicate statistically different groups determined by a post hoc Dunn's test



regression revealed significant variations in endophytic colonization depending on the plant segment and the fungal treatment (Table 3). Stems exhibited a notably higher likelihood of colonization than leaves (GLM:  $\chi^2 = 2.68$ ,  $p = 0.007$ ), suggesting a twofold increase in the likelihood of colonization. The fungal treatments had a significant effect: GHA significantly increased colonization compared with the Control (GLM:  $\chi^2 = 4.02$ ,  $p < 0.001$ ), while KA14 was even more effective (GLM:  $\chi^2 = 6.34$ ,  $p < 0.001$ ), i.e., around two times more effective than GHA. However, no significant difference was observed between roots and leaves (GLM:  $\chi^2 = -0.47$ ,  $p = 0.632$ ) or between days 7 and 14 after inoculation (GLM:  $\chi^2 = 0.43$ ,  $p = 0.664$ ), suggesting temporal stability of colonization.

**Table 3** Outputs of beta regression generalized linear model (GLM) summarizing the inoculation of entomopathogenic fungi in cabbage

	Estimate	Std. error	Z value	Pr(> z )
(Intercept)	-2.382	0.309	-7.701	< 0.001***
Root	-0.136	0.284	-0.479	0.632
Stem	0.780	0.290	2.687	0.007**
<i>B. bassiana</i> GHA	1.159	0.288	4.023	< 0.001***
<i>B. bassiana</i> KA14	1.917	0.302	6.344	< 0.001***
7 DAI	0.101	0.233	0.434	0.664

## Choice test assays

For the choice test performed with leaves harvested at 7 DAI, the tested aphids were more attracted to leaves from inoculated plants than to those of the controls. More aphids were observed on the discs treated with the *B. bassiana* GHA (GLMM:  $\chi^2_{(1)} = 15.13$ ,  $p < 0.001$ ) and *B. bassiana* KA14 (GLMM:  $\chi^2_{(1)} = 20.23$ ,  $p < 0.001$ ) than on the control discs. Interestingly, when the experiments were performed on leaves harvested 14 DAI, aphids showed no preference between fungal inoculated leaves (*B. bassiana* GHA or KA14) and control plants. Aphids' choice among treatments was not influenced by the time factor ( $p > 0.05$ ) (Fig. 4).

## Efficacy of *Beauveria bassiana* GHA treatment against *Brevicoryne brassicae* under field conditions

Significant differences were reported between the treatments in the “LRS” experiment ( $F_{(2,54)} = 64.33$ ,  $p < 0.001$ ) and the “SRS” experiment ( $F_{(2,54)} = 49.42$ ,  $p < 0.001$ ) for the infestation rate (Fig. 5A and B). Infestation rates were similar for all treatments in the first few days after transplanting (DAT). This was followed by a significantly higher number of aphids for the control plots until the end of the experiments ( $79.1 \pm 4.1\%$  and  $83.3 \pm 4.1\%$  at 63 DAT in the “LRS” and “SRS” experiments, respectively). In contrast, the Cyperscope 5EC and *B. bassiana* GHA treatments effectively reduced aphid infestation, and post hoc comparisons using Tukey's test indicated no significant difference among the two treatments.

The aphids' infestation on *B. brassicae* started at 14 and 7 DAT in the “LRS” and “SRS” experiments, respectively. About aphid number, significant differences were observed among the treatments in the “LRS” ( $F_{(2,54)} = 48.49$ ,  $p < 0.001$ ) and “SRS” ( $F_{(2,54)} = 75.67$ ,  $p < 0.001$ )

experiments. Over time, aphid numbers were stable in the plots treated with the fungus and chemical insecticide, compared with the control plots, where infestations continued to increase, reaching a maximum of  $123.6 \pm 23.1$  aphids at 63 DAT and  $146.6 \pm 17.6$  aphids at 56 DAT in “LRS” and “SRS” experiments, respectively. Post hoc comparisons using Tukey's test indicated that the EPF *B. bassiana* and the Cyperscope 5EC plots showed similar data compared to the control plots, which had the highest aphid number (Fig. 5C and D).

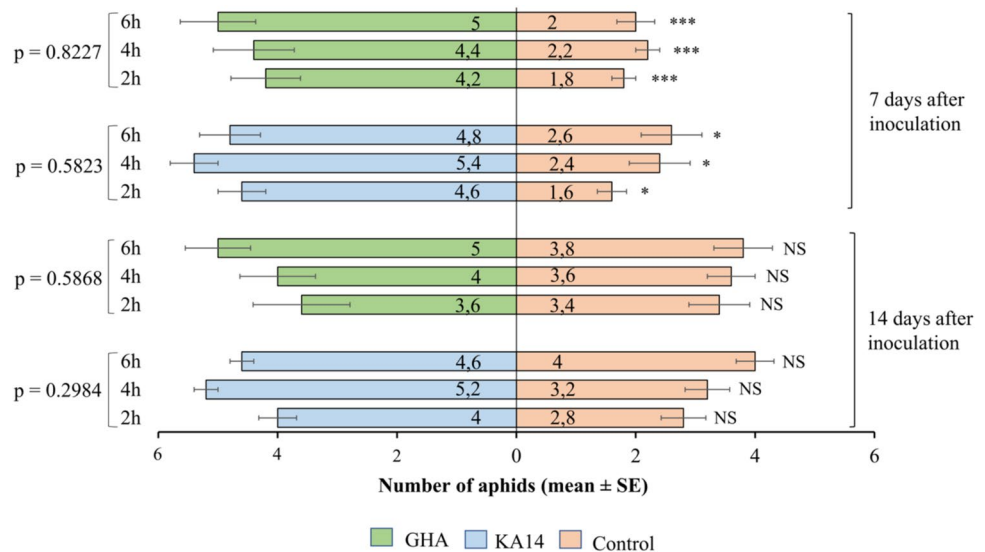
## Discussion

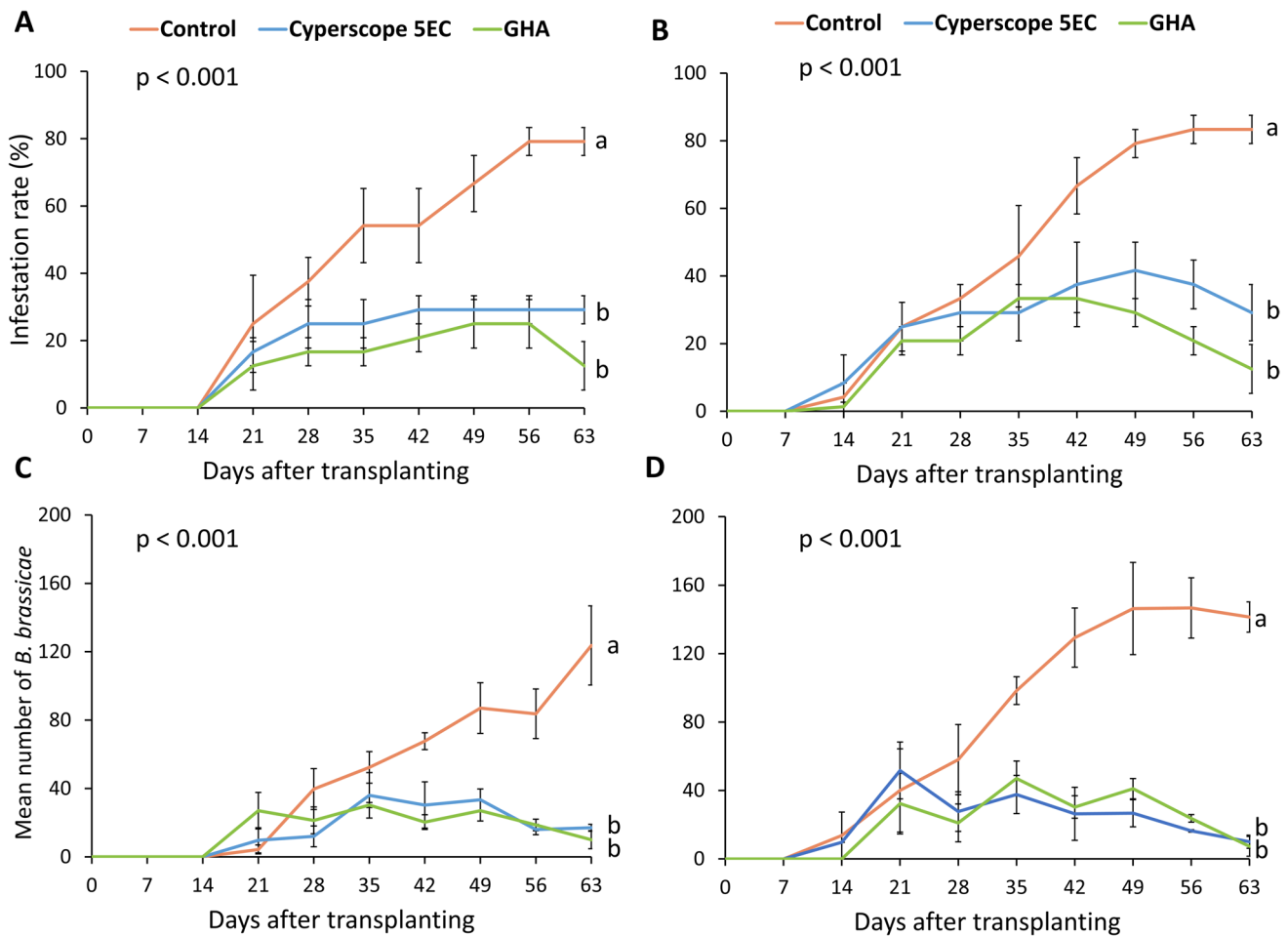
### Biocontrol potential of *Beauveria bassiana* under laboratory assays

#### Aphid mortality and survival probability

Survival probability revealed a significant effect of *B. bassiana* treatments on *B. brassicae* aphid mortality, whether by direct application or exposure to inoculated leaves. These results are consistent with previous studies demonstrating the pathogenicity of EPF *B. bassiana* against a wide range of insect pests. For example, Lugendo et al. (2025) evaluated the lethal and sublethal effects of *B. bassiana* (GHA) and *Metarhizium brunneum* (USDA 4556 and V275), both adjusted to  $1 \times 10^8$  conidia  $\text{mL}^{-1}$ , on the bean weevil *Bruchus rufimanus* (Boheman, 1833). They reported that *B. bassiana* GHA was the most effective strain, achieving an  $\text{LT}_{50}$  of 4 days and 96.6% mortality at 10 days. Similarly, Dessauvages et al. (2024) compared two *B. bassiana* strains (GHA and GxABT-1;  $1 \times 10^8$  conidia  $\text{mL}^{-1}$ ) in a direct spray experiment against the green peach aphid (*M. persicae*) reporting  $\text{LT}_{50}$  values of 5 and 6 days for GxABT-1

**Fig. 4** Selection of plants by aphids between Control and fungal inoculated plants (*Beauveria bassiana* GHA and KA14) in two-choice preference tests. For each test, 10 aphids were released and observed after 2, 4, and 6 h. On the left, the  $p$ -values represent tests comparing aphid distribution over time. On the right, there are leaves harvested 7 days and 14 days after inoculation of cabbage seedlings. Error bars are standard errors (SE)





**Fig. 5** Effects of *Beauveria bassiana* GHA (green line), Cyberscope 5EC (blue line), and Control (orange line) on the population of *Brevicoryne brassicae*. **A** Aphid infestation rate per plot during the long rainy season (LRS) experiment. **B** Aphid infestation rate per plot during the short rainy season (SRS) experiment. **C** Mean number

of *Brevicoryne brassicae* per plant during the LRS experiment. **D** Mean number of *Brevicoryne brassicae* during the SRS experiment. Error bars are standard errors (SE). Treatments followed by the same letter do not differ statistically at 5% significance according to the Tukey post hoc test

and GHA, respectively. The comparative pathogenicity of the commercial strain GHA and the indigenous strain KA14 highlights subtle but important differences in their speed of action. These differences may be due to genetic differences, the isolate virulence, the ability of spores to adhere to insect cuticles, or interactions with environmental factors such as humidity and temperature (Slowik et al. 2024; Oberti et al. 2025). These findings highlight the importance of selecting appropriate fungal strains to maximize efficacy.

Furthermore, the  $LT_{50}$  values observed with KA14 and GHA following exposure to treated leaves at both 7 DAI and 14 DAI support their efficacy against target pests over time. This finding suggests that *B. bassiana* remains effective after plant application, making it a viable biocontrol tool (Francis et al. 2022). In addition, these results indicate sustained fungal activity and potential compatibility with

cabbage as a host plant for endophytic colonization. In a related study, Qayyum et al. (2015) evaluated the pathogenicity of three *B. bassiana* strains against the tomato fruit worm (*H. armigera*) using inoculated tomato leaves. They found that *B. bassiana* WG-40 was the most effective strain, with leaf feeding resulting in the highest mortality 1 week after inoculation. However, this effect decreased significantly at 3 and 5 weeks after inoculation. Their findings suggest that the pathogenicity of *B. bassiana* is strongly influenced by the time elapsed since application. Further research is warranted to explore the mechanisms underlying isolate performance, particularly in plant-mediated applications, and to assess their long-term impact on pest populations and crop health.

### Endophytic potential of *Beauveria bassiana*

Our results suggest that *B. bassiana* could affect plant height, with differences observed at 14 DAI but not at 7 DAI. The lack of a significant effect at 7 DAI may be due to an insufficient incubation period for the endophyte to stimulate plant growth (Wang et al. 2024). Fungi require time to colonize plant tissues and modulate physiological processes such as nutrient uptake and phytohormone synthesis (Grabka et al. 2022). At 14 DAI, inoculated plants exhibited significantly higher normalized height increase values, likely due to improved colonization and increased metabolic activity of the fungal isolates. Endophytic EPF, including *B. bassiana*, are known to produce phytohormones such as indole-3-acetic acid (IAA), enhance nutrient uptake, and induce stress tolerance (Ownley et al. 2010; Dara et al. 2017; Ahsan et al. 2024). These findings highlight the importance of time as a critical factor in plant studies and emphasize the need for long-term monitoring to accurately evaluate the impact of these treatments on plant developmental trends. This is in accordance with Barra-Bucarei et al. (2020), who evaluated the effects of five *B. bassiana* strains on tomato growth parameters, including plant height, at 30 DAI. In their study, all strains resulted in numerically higher plant height than the Control, with the two best-performing strains, RGM-557 and RGM-731, achieving increases of 21% and 18%, respectively.

Our study highlights the performance of the endemic *B. bassiana* KA14, showing the highest and most consistent endophytism rates, particularly at 14 DAI, in comparison to the commercial strain GHA. A temporal decrease in colonization rates was observed for both treatments, which might be explained by the dynamic interactions between the endophytes and the host plant over time (de Carvalho et al. 2021). Notably, colonization was specific to tissues, with leaf and stem segments consistently showing higher endophytism rates than roots, irrespective of the treatment. These findings are consistent with those of Yogananda et al. (2023), who evaluated the colonization of cabbage tissues using different methods of inoculation (seed treatment, seedling root dip, soil drenching, and foliar spray). They reported that fungal colonization was higher in the inoculated cabbage tissues with limited movement to other plant tissues. In our study, foliar application may explain the higher emergence of the fungus in leaf tissues. These results suggest that tissue-specific factors and time-dependent dynamics are critical in shaping microbial colonization patterns (Yogananda et al. 2023). The observed colonization rates of *B. bassiana* strains in plant tissues indicate the compatibility of the fungus, particularly the indigenous isolate KA14, with cabbage plants, which holds promise for pest management. Further research is needed to elucidate the mechanisms driving these

colonization patterns and to optimize the use of high-performing strains in agricultural applications.

Aphid preference tests revealed a clear preference for *B. bassiana*-colonized leaves harvested at 7 DAI compared to non-fungal-treated leaves. However, no preference was observed at 14 DAI, suggesting a temporal change in the characteristics of the inoculated plants. The increased attractiveness of treated leaves at 7 DAI may be related to physiological or biochemical changes following fungal treatment, such as the production of volatile compounds induced by fungal infection. While these volatile compounds could play a role, this study did not measure them, and their role remains a putative perspective. Previous studies have demonstrated that plant- and fungus-derived volatile compounds, including aldehydes, terpenes, and other small molecules, can mediate insect behavior (González-Mas et al. 2019; Fingu-Mabola et al. 2020; Geedi et al. 2023). For example, Fingu-Mabola et al. (2020) reported that tobacco plants treated with *B. bassiana* were more attractive to *M. persicae* than uninoculated plants, possibly due to the high aldehyde production by fungus-inoculated plants. A study by Geedi et al. (2023) found that *M. persicae* was attracted to volatile compounds emitted by *B. bassiana* GHA, and this attraction was observed in both Petri dish and Y-tube olfactometer bioassays, indicating a significant behavioral response to fungal emissions. These results highlight the role of temporal dynamics in plant-insect-fungus interactions. In contrast, other studies have shown that fungus-inoculated plants deter phytophagous insects. For example, Dessauvages et al. (2024) found that sugar beet plants inoculated with *B. bassiana* GxABT-1 and GHA were less attractive to *M. persicae* than control plants. Future research should aim to investigate whether these preferences change under different environmental conditions or across different insect or host species, and to quantify the plant- and fungus-derived volatile cues underlying these interactions.

### In field experiments using *Beauveria bassiana* GHA

Both *B. bassiana* GHA and the chemical insecticide Cyper-scope 5EC significantly reduced *B. brassicae* infestation compared to the control plots. The observed effectiveness of *B. bassiana* is consistent with previous studies demonstrating the efficacy of this EPF in controlling a wide range of pest populations under field conditions, including *B. rufimanus* (Lugendo et al. 2025), fall armyworm *Spodoptera frugiperda* (J.E. Smith, 1797) (Kuzhuppillymyal-Prabhakarankutty et al. 2021), cotton aphid *Aphis gossypii* (Glover, 1877), strawberry aphid *Chaetosiphon fragaefolii* (Cockerell, 1901), and western flower thrip *Frankliniella occidentalis* (Pergande, 1895) (Mantzoukas et al. 2022). In another cabbage field experiment, Nouh et al. (2022) compared the efficacy of a commercial *B. bassiana* formulation with an

insecticide treatment. After weekly monitoring until harvest, they reported low *B. brassicae* infestations with aphids in the *B. bassiana*-treated plots, followed by chemically treated plots, while the highest infestations were observed in the control plots. The high-altitude region of Kivu where we carried out the experiments is known for its mild and cool climate, which may be favorable for fungal activity. The comparable efficacy of the fungal and chemical treatments suggests that *B. bassiana* could serve as a viable alternative or complement to chemical insecticides in IPM programs.

In terms of aphid number dynamics, the stability of *B. brassicae* infestations in treated plots suggests that both *B. bassiana* and Cyperscope 5EC were effective in suppressing early aphids' establishment. While Cyperscope 5EC, a synthetic pyrethroid, provides an immediate knockdown effect by inducing nerve paralysis in insect pests (Singh et al. 2022), *B. bassiana* acts more gradually by infecting and propagating within aphid populations, potentially reducing the risk of insect pest resurgence (Baverstock et al. 2010). In contrast, the rapid increase in aphid populations in control plots highlights the high reproductive potential of the pest and emphasizes the need for timely intervention (Munthali and Tshogofatso 2014). In this study, the greater efficacy of *B. bassiana* in the "LRS" experiment, conducted during the long rainy season, compared to the "SRS" experiment, conducted during the short rainy season, may be due to prevailing environmental conditions. Overall, the field efficacy of EPF, such as *B. bassiana*, is strongly influenced by abiotic factors such as rainfall, temperature, humidity, and exposure to sunlight (Jaronski 2010; Mweke et al. 2018). Overall, these findings support the great potential of *B. bassiana* as a biological control agent in different agroecosystems and under different environmental conditions, providing a sustainable alternative with a lower environmental impact than chemical insecticides.

## Conclusion

Exploring the potential of introduced or native insect pathogenic fungi in different agricultural and environmental contexts is of utmost interest to select novel and more eco-friendly biocontrol agents that can support modern agriculture. Laboratory results showed that the endemic strain *B. bassiana* KA14 has promising entomopathogenic and endophytic potential compared to the commercial strain *B. bassiana* GHA. These results support the integration of *B. bassiana* into sustainable pest management strategies that reduce reliance on chemical insecticides. However, more in-depth investigation should be conducted to explore the long-term sustainability of *B. bassiana* applications and its potential synergistic effects when combined with other IPM strategies.

**Acknowledgements** We are grateful to Emilie Bera for the technical assistance during laboratory investigations. We extend our thanks to Arnaud Segers for sharing the Botanigard 22WP for field application. The authors also thank local collaborators who assisted with the field trials in Eastern DRC.

**Author Contribution** Conceptualization: P.N.G. and F.F., methodology: I.B.F. and F.F., investigation and data collection: P.N.G., data analysis: P.N.G. and M.C.C., writing—original draft preparation: P.N.G., writing—review and editing: M.C.C., K.D., J-P.M.M., F.M.H., R.C.M., F.F., and I.B.F., funding acquisition: P.N.G. and F.F. All authors read and approved the final manuscript.

**Funding** This research was funded by the Belgian Development Agency «Enabel» within PRECOB (Programme de Renforcement des Capacités par l'Octroi des bourses).

## Declarations

**Ethical Approval** Not applicable.

**Conflict of interest** I.B.F. is an associate editor of the journal Neotropical Entomology, and her position does not interfere with any aspect of the publication process.

## References

- Ahsan SM, Injamum-Ul-Hoque Md, Das AK, Rahman MdM, Mollah MdMI, Paul NC, Choi HW (2024) Plant-entomopathogenic fungi interaction: recent progress and future prospects on endophytism-mediated growth promotion and biocontrol. *Plants* 13:1420. <https://doi.org/10.3390/plants13101420>
- Altaf N, Ullah MI, Afzal M, Arshad M, Ali S, Rizwan M, Al-Shuraym LA, Alhelaify SS, Sayed S (2023) Endophytic colonization by *Beauveria bassiana* and *Metarhizium anisopliae* in maize plants affects the fitness of *Spodoptera frugiperda* (Lepidoptera: Noctuidae). *Microorganisms* 11:1067. <https://doi.org/10.3390/microorganisms11041067>
- Arinanto LS, Hoffmann AA, Ross PA, Gu X (2024) Hormetic effect induced by *Beauveria bassiana* in *Myzus persicae*. *Pest Manag Sci* 80:3726–3733. <https://doi.org/10.1002/ps.8075>
- Balasha AM, Nsele MK (2019) Pesticide use practices by Chinese cabbage growers in suburban environment of Lubumbashi (DR Congo): main pests, costs and risks. 56–64. <https://doi.org/10.12691/jaaepa-2-1-8>
- Barra-Bucarei L, González MG, Iglesias AF, Aguayo GS, Peñalosa MG, Vera PV (2020) *Beauveria bassiana* multifunction as an endophyte: growth promotion and biologic control of *Trialeurodes vaporariorum*, (Westwood) (Hemiptera: Aleyrodidae) in tomato. *InSects* 11:591. <https://doi.org/10.3390/insects11090591>
- Baverstock J, Roy HE, Pell JK (2010) Entomopathogenic fungi and insect behaviour: from unsuspecting hosts to targeted vectors. *Bio-Control* 55:89–102. <https://doi.org/10.1007/s10526-009-9238-5>
- Ben Fekih I, Jensen AB, Boukhris-Bouhachem S, Pozsgai G, Rezgui S, Rensing C, Eilenberg J (2019) Virulence of two Entomophthoralean fungi, *Pandora neoaphidis* and *Entomophthora planchoniana*, to their conspecific (*Sitobion avenae*) and heterospecific (*Rhopalosiphum padi*) aphid hosts. *InSects* 10:54. <https://doi.org/10.3390/insects10020054>
- Biryol S, Demirbağ Z, Erdoğan P, Demir I (2022) Development of *Beauveria bassiana* (Ascomycota: Hypocreales) as a mycoinsecticide to control green peach aphid, *Myzus persicae* (Homoptera:

- Aphididae) and investigation of its biocontrol potential. *J Asia-Pac Entomol* 25:101878. <https://doi.org/10.1016/j.aspen.2022.101878>
- Certis (2021) Fiche étiquette Botanigard 22WP. <https://www.certiseuro.pe/nos-produits/produit/detail/botanigardr-22-wp>. Accessed 17 Oct 2022
- Cokola MC, Ben Fekih I, Bisimwa EB, Caparros Megido R, Delvigne F, Francis F (2023) Natural occurrence of *Beauveria bassiana* (Ascomycota: Hypocreales) infecting *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) and earwig in eastern DR Congo. *Egypt J Biol Pest Control* 33:54. <https://doi.org/10.1186/s41938-023-00702-2>
- Dara SK, Dara SSR, Dara SS (2017) Impact of entomopathogenic fungi on the growth, development, and health of cabbage growing under water stress. *Am J Plant Sci* 08:1224–1233. <https://doi.org/10.4236/ajps.2017.86081>
- de Carvalho CR, Ferreira MC, Rosa LH (2021) Ecology of neotropical endophytic fungi. In: Rosa LH (ed) Neotropical endophytic fungi: diversity, ecology, and biotechnological applications. Springer International Publishing, Cham, pp 1–9
- Dessauvages K, Scheiffer M, Francis F, Ben Fekih I (2024) A new isolate *Beauveria bassiana* GxABT-1: efficacy against *Myzus persicae* and promising impact on the beet mild yellow virus-aphid association. *InSects* 15:697. <https://doi.org/10.3390/insects15090697>
- Fingu-Mabola JC, Martin C, Bawin T, Verheggen FJ, Francis F (2020) Does the infectious status of aphids influence their preference towards healthy, virus-infected and endophytically colonized plants? *InSects* 11:435. <https://doi.org/10.3390/insects11070435>
- Francis F, Fingu-Mabola JC, Ben Fekih I (2022) Direct and endophytic effects of fungal entomopathogens for sustainable aphid control: a review. *Agriculture* 12:2081. <https://doi.org/10.3390/agriculture12122081>
- Gakuru PN, Noël G, Muhashy HF, Francis F (2024) Monitoring entomofauna associated with cabbage, *Brassica oleracea* L. var capitata in eastern DR Congo. *J Insect Biodivers Syst* 10:453–466. <https://doi.org/10.61186/jibs.10.3.453>
- Geedi R, Canas L, Reding ME, Ranger CM (2023) Attraction of *Myzus persicae* (Hemiptera: Aphididae) to volatiles emitted from the entomopathogenic fungus *Beauveria bassiana*. *Environ Entomol* 52:31–38. <https://doi.org/10.1093/ee/nvac100>
- González-Mas N, Sánchez-Ortiz A, Valverde-García P, Quesada-Moraga E (2019) Effects of endophytic entomopathogenic ascomycetes on the life-history traits of *Aphis gossypii* Glover and its interactions with melon plants. *InSects* 10:165. <https://doi.org/10.3390/insects10060165>
- Grabka R, d'Entremont TW, Adams SJ, Walker AK, Tanney JB, Abbasi PA, Ali S (2022) Fungal endophytes and their role in agricultural plant protection against pests and pathogens. *Plants* 11:384. <https://doi.org/10.3390/plants11030384>
- Hao Z-P, Zhan H-X, Wang Y-L, Hou S-M (2019) How cabbage aphids *Brevicoryne brassicae* (L.) make a choice to feed on *Brassica napus* cultivars. *InSects* 10:75. <https://doi.org/10.3390/insects10030075>
- Jaber LR, Enkerli J (2017) Fungal entomopathogens as endophytes: can they promote plant growth? *Biocontrol Sci Technol* 27:28–41. <https://doi.org/10.1080/09583157.2016.1243227>
- Jaber LR, Araj S-E, Qasem JR (2018) Compatibility of endophytic fungal entomopathogens with plant extracts for the management of sweetpotato whitefly *Bemisia tabaci* Gennadius (Homoptera: Aleyrodidae). *Biol Control* 117:164–171. <https://doi.org/10.1016/j.biocontrol.2017.11.009>
- Jaronski ST (2010) Ecological factors in the inundative use of fungal entomopathogens. *Biocontrol* 55:159–185. <https://doi.org/10.1007/s10526-009-9248-3>
- Kassambara A, Kosinski M, Bieчек P (2016) Survminer: drawing survival curves using “ggplot2”. R package version 0.4.9. <https://CRAN.R-project.org/package=survminer>
- Kuzhuppillymyal-Prabhakaranakutty L, Ferrara-Rivero FH, Tamez-Guerra P, Gomez-Flores R, Rodríguez-Padilla MC, Ek-Ramos MJ (2021) Effect of *Beauveria bassiana*-seed treatment on *Zea mays* L. response against *Spodoptera frugiperda*. *Appl Sci*. <https://doi.org/10.3390/app11072887>
- Lugendo AR, Ben Fekih I, Caparros Megido R, Pierreux J, Francis F, Segers A (2025) Efficacy of entomopathogenic fungi against *Bruchus rufimanus* (coleoptera: chrysomelidae) in laboratory and field trials using droplet spraying technique. *Agriculture (Basel)* 15:376. <https://doi.org/10.3390/agriculture15040376>
- Mantzoukas S, Daskalaki E, Kitsiou F, Papantzikos V, Servis D, Bitivanos S, Patakioutas G, Eliopoulos PA (2022) Dual action of *Beauveria bassiana* (Hypocreales; Cordycipitaceae) endophytic stains as biocontrol agents against sucking pests and plant growth biostimulants on melon and strawberry field plants. *Microorganisms* 10:2306
- Mascarin GM, Jaronski ST (2016) The production and uses of *Beauveria bassiana* as a microbial insecticide. *World J Microbiol Biotechnol* 32:177. <https://doi.org/10.1007/s11274-016-2131-3>
- Mireles-Valdez JS, Sandoval-Coronado CF, Blanco MaGM, Elias-Santos M, Rojas-Verde MaG, Rodriguez-Castro A (2023) Effect of *Beauveria bassiana* 1 Conidia against larval stages of *Trichoplusia ni* 2. *Southwest Entomol* 48. <https://doi.org/10.3958/059.048.0411>
- Mpumi N, Machunda RS, Mtei KM, Ndakidemi PA (2020) Selected insect pests of economic importance to *Brassica oleracea*, their control strategies and the potential threat to environmental pollution in Africa. *Sustainability* 12:3824. <https://doi.org/10.3390/su12093824>
- Munthali DC, Tshogafatso AB (2014) Factors affecting abundance and damage caused by cabbage aphid, *Brevicoryne brassicae* on four *Brassica* leafy vegetables: *Brassica oleracea* var. *Acephala*, *B. chinense*, *B. napus* and *B. carinata*. *Open Entomol J* 8:1–9. <https://doi.org/10.2174/1874407901408010001>
- Mweke A, Ulrichs C, Nana P, Akutse KS, Fiaboe KKM, Maniania NK, Ekesi S (2018) Evaluation of the entomopathogenic fungi *Metarhizium anisopliae*, *Beauveria bassiana* and *Isaria* sp. for the management of *Aphis craccivora* (Hemiptera: Aphididae). *J Econ Entomol* 111:1587–1594. <https://doi.org/10.1093/jee/toy135>
- Nouh G, Eid A, Kamel H, Adly D (2022) Efficacy of the entomopathogenic fungus, *Beauveria bassiana* and certain insecticides on some cabbage insects in field. *Egypt J Agric Res* 100:499–510. <https://doi.org/10.21608/ejar.2022.149858.1249>
- Ntow WJ, Gijzen HJ, Kelderman P, Drechsel P (2006) Farmer perceptions and pesticide use practices in vegetable production in Ghana. *Pest Manag Sci* 62:356–365. <https://doi.org/10.1002/ps.1178>
- Oberti H, Sessa L, Oliveira-Rizzo C, Di Paolo A, Sanchez-Vallet A, Seidl MF, Abreo E (2025) Novel genomic features in entomopathogenic fungus *Beauveria bassiana* ILB308: accessory genomic regions and putative virulence genes involved in the infection process of soybean pest *Piezodorus guildinii*. *Pest Manag Sci* 81:2323–2336. <https://doi.org/10.1002/ps.8631>
- Ownley BH, Gwinn KD, Vega FE (2010) Endophytic fungal entomopathogens with activity against plant pathogens: ecology and evolution. *Biocontrol* 55:113–128. <https://doi.org/10.1007/s10526-009-9241-x>
- Parsa S, Ortiz V, Vega FE (2013) Establishing fungal entomopathogens as endophytes: towards endophytic biological control. *J Vis Exp* 50360. <https://doi.org/10.3791/50360>
- Pereira AJ, Cardoso IM, Araújo HD, Santana FC, Carneiro APS, Coelho SP, Pereira FJ (2019) Control of *Brevicoryne brassicae* (Hemiptera: Aphididae) with extracts of *Agave americana*

- var. *Marginata* Trel. in *Brassica oleracea* crops. Ann Appl Biol 174:14–19. <https://doi.org/10.1111/aab.12471>
- Prabha S, Yadav A, Kumar A, Yadav A, Yadav HK, Kumar S, Yadav RS, Kumar R (2016) Biopesticides - an alternative and eco-friendly source for the control of pests in agricultural crops. Plant Arch 16:902–906
- Qayyum MA, Wakil W, Arif MJ, Sahi ST, Dunlap CA (2015) Infection of *Helicoverpa armigera* by endophytic *Beauveria bassiana* colonizing tomato plants. Biol Control 90:200–207. <https://doi.org/10.1016/j.biocontrol.2015.04.005>
- R Core Team (2024) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.r-project.org/>
- Saha T, Kalmesh M, Nithya C, Singh MP, Kumari K (2018) Insect pest management of preharvest vegetables for better postharvest quality. In: Siddiqui MW (ed) Preharvest modulation of postharvest fruit and vegetable quality. Academic Press, Oxford, pp 277–300
- Shang J, Hong S, Wang C (2024) Fights on the surface prior to fungal invasion of insects. PLoS Pathog 20:e1011994. <https://doi.org/10.1371/journal.ppat.1011994>
- Shehzad M, Tariq M, Ali Q, Aslam A, Mukhtar T, Akhtar MF, Gulzar A, Faisal M (2022) Evaluation of insecticidal activity of *Beauveria bassiana* against different instar larvae of *Plutella xylostella* by using two different methods of application. Int J Trop Insect Sci 42:1471–1476. <https://doi.org/10.1007/s42690-021-00665-7>
- Shonga E, Getu E (2021) Population dynamics of cabbage aphid, *Brevicoryne brassicae* L. (Homoptera : Aphididae) in relation to weather factors on major brassica crops in central rift valley of Ethiopia : baseline studies for the management of the pest. Int J Trop Insect 41:455–462
- Singh A, Singh A, Singh P, Chakravarty A, Singh A, Singh P, Mishra MK, Singh V, Srivastava AK, Aggarwal H, Sagadevan S (2022) Insecticidal activity, toxicity, resistance and metabolism of pyrethroids: a review. Sci Technol Indones 7:238–250. <https://doi.org/10.26554/sti.2022.7.2.238-250>
- Slowik AR, Hesketh H, Sait SM, De Fine Licht HH (2024) Thermal ecology shapes disease outcomes of entomopathogenic fungi infecting warm-adapted insects. J Invertebr Pathol 204:108106. <https://doi.org/10.1016/j.jip.2024.108106>
- Smithson M, Verkuilen J (2006) A better lemon squeezer? Maximum-likelihood regression with beta-distributed dependent variables. Psychol Methods 11:54–71. <https://doi.org/10.1037/1082-989X.11.1.54>
- Świergiel W, Meyling NV, Porcel M, Rämert B (2016) Soil application of *Beauveria bassiana* GHA against apple sawfly, *Hoplocampa testudinea* (Hymenoptera: Tenthredinidae): field mortality and fungal persistence. Insect Sci 23:854–868. <https://doi.org/10.1111/1744-7917.12233>
- Therneau T (2024) A package for survival analysis in R. <https://doi.org/10.32614/CRAN.package.survival>
- Torkaman Z, Talaei-Hassanloui R, Khorramnejad A, Pashaei M (2023) Effects of endophytism by *Beauveria bassiana* (Cordycipitaceae) on plant growth, *Fusarium* (Nectriaceae) disease, and Sunn pest *Eurygaster integriceps* (Hemiptera: Scutelleridae) in wheat (Poaceae). Can Entomol 155. <https://doi.org/10.4039/tce.2022.43>
- Tudi M, Daniel Ruan H, Wang L, Lyu J, Sadler R, Connell D, Chu C, Phung DT (2021) Agriculture development, pesticide application and its impact on the environment. Int J Environ Res Public Health 18:1112. <https://doi.org/10.3390/ijerph18031112>
- Vu VH, Hong SI, Kim K (2007) Selection of entomopathogenic fungi for aphid control. J Biosci Bioeng 104:498–505. <https://doi.org/10.1263/jbb.104.498>
- Wang Q, Zhang X, Xie Q, Tao J, Jia Y, Xiao Y, Tang Z, Li Q, Yuan M, Bu T (2024) Exploring plant growth-promoting traits of endophytic fungi isolated from *Ligusticum chuanxiong* Hort and their interaction in plant growth and development. J Fungi 10:713. <https://doi.org/10.3390/jof10100713>
- Wickham H (2016) Ggplot2: elegant graphics for data analysis, second edition. Springer International Publishing, Houston
- Yogananda T, Ramanagouda SH, Venkateshalu B, Jamunarani GS, Rashmi SH, Awati M, Hadimani HP (2023) Colonization and endophytic effect of *Beauveria bassiana* (Bals.-Criv.) Vuill. UHSB-END1 against *Myzus persicae* (Sulzer) and *Plutella xylostella* (L.) in cabbage. Egypt J Biol Pest Control 33:47. <https://doi.org/10.1186/s41938-023-00698-9>

**Publisher's Note** Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Springer Nature or its licensor (e.g. a society or other partner) holds exclusive rights to this article under a publishing agreement with the author(s) or other rightsholder(s); author self-archiving of the accepted manuscript version of this article is solely governed by the terms of such publishing agreement and applicable law.