

## ORIGINAL ARTICLE

## Evaluating bacteria, fungi and viruses for controlling shoot borer (*Cryptophlebia ombrodelta*) damage in *Erythrophleum fordii* plantations in Vietnam

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DOI: 10.24425/jppr.2026.2966

Received: March 28, 2025

Accepted: August 12, 2025

Online publication: August 31, 2026

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Responsible Editor:  
Sándor Keszthelyi

### Abstract

*Erythrophleum fordii* has been prioritized for use in sawtimber production and forest restoration programs with native tree species in Vietnam. However, the shoot borer *Cryptophlebia ombrodelta* (Lower, 1898) (Lepidoptera: Tortricidae) is becoming a serious pest in *E. fordii* plantations. This study evaluated the potency of biological agents to control *C. ombrodelta* in laboratory and field experiments. Eight biological control agents (BCAs) were prescreened in a laboratory trial and the five most effective BCAs were evaluated in a one-year-old plantation. Spraying foliage with *Bacillus thuringiensis* var. *kurstaki* + granulosis virus or nuclear polyhedrosis virus at concentrations of 4 g · l<sup>-1</sup> and 4 ml · l<sup>-1</sup>, respectively, reduced damage from *C. ombrodelta* by over 70% compared to the water control. The findings can guide further research on the timing and frequency of applications of BCAs necessary to manage the pest in young stands of *E. fordii*.

**Keywords:** biological control, insect pest, ironwood, pest management, plantation forestry

## Introduction

*Erythrophleum fordii* Oliv. (ironwood) is native to southern China and southeast Asia (Sein and Mitlöhner 2011). It is widely cultivated in China (Sein and Mitlöhner 2011; Zhao *et al.* 2021) and in parts of northern and central Vietnam (Sein and Mitlöhner 2011). The tree is renowned for its high quality timber (Sein and Mitlöhner 2011), and as a source of pharmaceutical compounds (Vo *et al.* 2021). This species has been endorsed for reforestation in Vietnam since 2014 (Hai *et al.* 2023). Nevertheless, in northern Vietnam the success of commercial plantings is being undermined by widespread damage from the shoot borer *Cryptophlebia ombrodelta* (Lower, 1898) (Lepidoptera: Tortricidae) (Nguyen *et al.* 2023).

*Cryptophlebia ombrodelta* is widely distributed in parts of Asia and Oceania extending from India to Hawaii (Brown *et al.* 2019; Patel *et al.* 2023). It attacks fruit and seeds of horticultural trees such as macadamia, litchi and longan (Jones 1995), and it can be a stem borer in a range of other trees in Fabaceae and Sapindaceae (Gupta *et al.* 2017; Patel *et al.* 2023). Also, this pest has been reported to attack shoots of *E. fordii* in China (Zhao *et al.* 2013).

There is considerable potential to apply biological control agents (BCAs) against pests in forest ecosystems (Balla *et al.* 2021; Fischbein and Corley 2022; Budziszewska and Bereś 2024), and the Vietnamese government has developed policies to promote the

use of BCAs in plantation forestry (Hai *et al.* 2023). Globally, there has been significant progress in the application of entomopathogenic BCAs in commercial crops (Arthurs and Dara 2019; Hatting *et al.* 2019), including entomopathogenic bacteria (Sezen *et al.* 2007; Chi *et al.* 2025), fungi (Sezen *et al.* 2007; Jaber and Ownley 2017; Chi and Anh 2025) and viruses (Moore 2002). Furthermore, an alphabaculovirus was effective against *Cryptophlebia peltastica* (Meyrick, 1921) (Lepidoptera: Tortricidae) (Marsberg *et al.* 2018). However, there are no studies on the management of *C. ombrodelta* in forest plantations. Therefore, this study was undertaken to screen BCAs available in Vietnam for the management of *C. ombrodelta* in young *E. fordii* plantations.

## Materials and Methods

### Laboratory experiment

To identify biocontrol agents with potential to kill *C. ombrodelta* larvae, a laboratory experiment was set up in a fully randomized design comprising eight biocontrol agents (*Bacillus bombysepticus*, *B. thuringiensis* var. *kurstaki*, *B. thuringiensis* var. *kurstaki* + granulosis virus, *Beauveria bassiana*, *B. bassiana* B2, *Metarhizium anisopliae*, nuclear polyhedrosis virus and *Serratia marcescens*) (Table 1) and a sterilized water control x 3 spray rates (2, 4 and 6 g · l<sup>-1</sup> or 2, 4 and 6 ml · l<sup>-1</sup>) of the stock concentrations recommended by the manufacturers (Table 1) x 4 replicates. Each replicate had 9 *C. ombrodelta* larvae.

*Cryptophlebia ombrodelta* was mass-reared in a laboratory on artificial food developed by Zhao *et al.* (2019) containing corn powder (18.95%), soybean

powder (18.95%), yeast powder (7.58%), glucose (3.79%), casein (2.53%), ascorbic acid (0.76%), methyl nipagin (0.51%), sorbic acid (0.25%), cholesterol (0.05%), tomato ketchup (37.91%), dried *Erythrophleum fordii* branch powder (5.05%), agar (3.54%) and formaldehyde (0.13%). Third instar larvae with lengths of 11–12 mm and diameters of 2.2–2.5 mm were used for this experiment. Shoots of *E. fordii* were sourced from two-year-old plantations in Nam Son commune, Soc Son district, Hanoi that were without damage from *C. ombrodelta*. Shoots with diameters of 0.8–0.9 cm were cut into 20 cm lengths, and placed vertically in 10 × 10 × 10 cm plastic containers containing about 3 cm of water. Holes were made to allow larvae to enter the stem as quickly as possible in a uniform location. An electric drill was used to bore a 0.3 cm diameter sloping hole, 1.2–1.5 cm deep in each stem, 5–6 cm below the apical bud. The treatments were sprayed onto the 3rd instar larvae at a dosage of 0.2 ml · larva<sup>-1</sup> using a mini spray bottle (10 ml) and one larva was then released into each hole. Each larva was transferred with a soft bristle brush to the shoot so that the head was facing the hole entrance, and entry then occurred without interference. The shoots of each treatment were sprayed at a dosage of 1.0 ml · shoot<sup>-1</sup> with the same biocontrol agent that was used on the larvae. To enhance surface spread and adhesion, treatments were mixed with 1% Tween 80 prior to spraying. After spraying, the shoots were placed in individual netted cages (30 × 20 × 20 cm), and maintained at 25–27°C and 65–68% humidity on laboratory benches. The number of parasitized larvae, determined by their color change, and dead larvae were recorded after 7 days. This time was chosen based on preliminary trials in which the time to death was monitored.

**Table 1.** Biological agents tested for control of *Cryptophlebia ombrodelta*

| Active ingredient                                               | Source                                                                                                                | Stock concentration*                          |
|-----------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------|-----------------------------------------------|
| <i>Bacillus bombysepticus</i>                                   | Entomopathogenic bacterium (strain FPRC23) isolated from <i>Ceracris kiangsu</i> nymph (Chi <i>et al.</i> 2025)       | 10 <sup>8</sup> CFU · ml <sup>-1</sup>        |
| <i>B. thuringiensis</i> var. <i>kurstaki</i>                    | Delfin 32WG (SDS Biothech K.K., Tokyo, Japan)                                                                         | 32 BIU · g <sup>-1</sup>                      |
| <i>B. thuringiensis</i> var. <i>kurstaki</i> + granulosis virus | Bitadin WP (Nong Sinh Co. Ltd., Hanoi, Vietnam)                                                                       | 16 IU + 10 <sup>5</sup> PIB · g <sup>-1</sup> |
| <i>Beauveria bassiana</i>                                       | Muskardin (CPC Co. Ltd., Can Tho, Vietnam)                                                                            | 10 <sup>8</sup> CFU · g <sup>-1</sup>         |
| <i>B. bassiana</i> B2                                           | Entomopathogenic fungal (strain B2) isolated from <i>Endocrita vietnamensis</i> larva (Chi and Anh 2025)              | 10 <sup>8</sup> CFU · ml <sup>-1</sup>        |
| <i>Metarhizium anisopliae</i>                                   | Metarhizium (META Co. Ltd., Ho Chi Minh, Vietnam)                                                                     | 10 <sup>3</sup> CFU · g <sup>-1</sup>         |
| Nuclear polyhedrosis virus                                      | Heli-cide (Pest Control Pvt. Ltd., Mumbai, India)                                                                     | 10 <sup>9</sup> CFU · ml <sup>-1</sup>        |
| <i>Serratia marcescens</i>                                      | Entomopathogenic bacterium (strain FPRC14) isolated from <i>Episparis tortuosalis</i> larva (Long <i>et al.</i> 2023) | 10 <sup>8</sup> CFU · ml <sup>-1</sup>        |

\*recommended by the manufacturer

## Field trial

The five most effective BCAs (*Bacillus bombysepticus*, *B. thuringiensis* var. *kurstaki*, *B. thuringiensis* var. *kurstaki* + granulosis virus, nuclear polyhedrosis virus and *Serratia marcescens*) in the laboratory experiment were chosen for evaluation in the field. The control treatment was clean water. *Erythrophleum fordii* plantations were surveyed in Tam Dao district, Vinh Phuc province (21.501127 N, 105.547417 E, 206 m a.s.l.) and a 13-ha compartment was chosen in April 2023 for the experiment with about 52% damage incidence from *C. ombrodelta*. The plantation was established in March 2022 using one-year-old seedlings, at a density of 1,660 trees · ha<sup>-1</sup>, with 2 m between trees and 3 m within rows. The seed was obtained from selected mother trees in Vinh Phuc province. Planting holes (30 × 30 × 30 cm) received 200 g of 5 : 10 : 3 NPK fertilizer before backfilling, and weeds were cut by hand every six months.

A randomized block design experiment, comprised of four blocks, was set up. Each block contained six plots (500 m<sup>2</sup>), and each plot had 80 trees. The treatments were randomly ascribed to the plots. The plots were separated by at least 20 m buffers that were left untreated. The BCAs were used at concentrations of 4 g · l<sup>-1</sup> or 4 ml · l<sup>-1</sup> as follows: 4 × 10<sup>8</sup> CFU · l<sup>-1</sup> *B. bombysepticus*, 128 BIU/l *B. thuringiensis* var. *kurstaki*, 64 IU *B. thuringiensis* var. *kurstaki* + 4 × 10<sup>5</sup> PIB · l<sup>-1</sup> granulosis virus, 4 × 10<sup>9</sup> CFU · l<sup>-1</sup> nuclear polyhedrosis virus, and 4 × 10<sup>8</sup> CFU · l<sup>-1</sup> (*S. marcescens*) to each tree. All treatments were mixed with 1% Tween 80. The treatments were sprayed to the shoots and foliage using an electric sprayer at the rate of 0.5 l · tree<sup>-1</sup>, which was enough to wet the entire tree. The treatments were applied over 1–2 hours in the late afternoon, without rain. Treatments were applied twice, 15 days apart, in April 2023. Repeat spraying after 14–15 days is recommended to ensure effective control when using BCAs (Chi *et al.* 2021a; Chi *et al.* 2023).

The trees were assessed at the time the trial was established (April 11, 2023) and again after 30 days (May 11, 2023) following the method of Nguyen *et al.* (2023). The following parameters were observed: the presence of *C. ombrodelta* excrement/exudation at the shoot, and plant recovery from damage. Damage at the time the trial was established was assessed as follows: 0 – no stem lesion, no excrement, healthy shoot; 1 – length of stem lesion <5 cm with new excrement; 2 – length of stem lesion 5 to <10 cm with new excrement; 3 – length of stem lesion 10 to <15 cm with new excrement, wilted shoot; and 4 – length of stem lesion >15 cm with abundant new excrement, dead shoot. The damage at the end of the trial was assessed at five levels, where: 0 – healthy tree, old lesion (if present) without excrement, one vigorous new shoot; 1 – length

of lesion <5 cm with new excrement, 1 new shoot; 2 – length of lesion 5 cm to <10 cm with new excrement, 2 new shoots; 3 – length of lesion 10 cm to <15 cm with new excrement, 3 new shoots, wilted shoots; 4 – length of the lesion >15 cm with much new excrement, dead main shoot.

## Data analysis

From the number of parasitized or dead larvae, the percentage of parasitized or dead larvae (DP%) was calculated using equation 1 (Chi and Anh 2025):

$$DP\% = (n/N) \times 100, \quad (1)$$

where:

n – the number of parasitized or dead larvae;  
N – the total number of larvae.

From the results of damage classification, the damage incidence (P%) was calculated using equation 2 (Chi *et al.* 2021b):

$$P\% = (n/N) \times 100, \quad (2)$$

where:

n – the number of trees attacked by *C. ombrodelta*;  
N – the total number of trees assessed.

The average damage index (DI) in each plot was calculated using equation 3 (Chi *et al.* 2021b):

$$DI = (\sum n_i \times v_i) / N, \quad (3)$$

where:

n<sub>i</sub> – the number of infected trees at damage index i;  
v<sub>i</sub> – the damage index at level i;  
N – the total number of trees assessed.

The corrected efficacy of each biocontrol agent in the field trial was calculated according to the following equation (4) of Henderson and Tilton (1955):

$$E = (1 - (C_b \times T_a) / (C_a \times T_b)) \times 100, \quad (4)$$

where:

C<sub>b</sub> – the damage incidence/damage index of the control treatment at the start of the trial;  
T<sub>b</sub> – the damage incidence/damage index of the biocontrol agent at the start of the trial;  
C<sub>a</sub> – the damage incidence/damage index of the control treatment at the end of the trial;  
T<sub>a</sub> – the damage incidence/damage index of the biocontrol agent at the end of the trial.

Plot mean values for the percentage of parasitized or dead larvae, P% and DI were calculated, and the block and plot mean data were analyzed using GenStat Release 12.1 software package (VSN International Ltd., Hemel Hempstead, UK). The Kolmogorov-Smirnov Test was used for testing data distribution. The percentage of parasitized or dead larvae, the damage

index and damage incidence were log-transformed before analysis. Significant control effectiveness of biocontrol agents against *C. ombrodelta* at different concentrations in the laboratory experiment were analyzed with two-way ANOVA (biocontrol agent  $\times$  concentration), followed by Tukey's Multiple Range Test for comparisons of means. The effect of biological control treatments on damage by *C. ombrodelta* in the field trial was tested using one-way analysis of variance (ANOVA), followed by Tukey's Multiple Range Test (using GenStat 12.1 software).

## Results

### Laboratory experiment

Two-factor analysis of variance (biocontrol agent  $\times$  concentration) showed a significant difference in the percentage of parasitized and dead larvae between biocontrol agents (Table 2) and concentration ( $p < 0.05$ ), but there was no interaction between the factors for the percentage of parasitized larvae ( $p = 0.802$ ) or the percentage of dead larvae ( $p = 0.818$ ).

In this experiment, *B. thuringiensis* var. *kurstaki* + granulosis virus (Fig. 1g) and the nuclear polyhedrosis virus (Fig. 1f) had the strongest parasitic effect on *C. ombrodelta*, with the percentage of parasitized larvae exceeding 70% and the percentage of dead larvae over 60%. The three entomopathogenic fungi *Beauveria bassiana* (Fig. 1i), *B. bassiana* B2 and *Metarhizium anisopliae* treatments had low rates of parasitized (41.7–48.2%) and dead (37.0–42.6%) larvae. In the control treatment (sterilized water), all the larvae were alive after 7 days (Table 2). Furthermore, the treatment rates of 4 and 6 g  $\cdot$  l<sup>-1</sup> or 4 and 6 ml  $\cdot$  l<sup>-1</sup> resulted in higher numbers ( $p = 0.03$ ) of parasitized and dead larvae than 2 g  $\cdot$  l<sup>-1</sup> or 2 ml  $\cdot$  l<sup>-1</sup> (data not shown).

The one-way analysis results of dosage rates for each BCA showed that the percentage of parasitized larvae was significantly different ( $p = 0.016$ – $0.020$ ) for *B. bombysepticus*, *B. thuringiensis* var. *kurstaki* and *S. marcescens*. Furthermore, the percentage of dead larvae was significantly different ( $p = 0.003$ – $0.025$ ) for *B. bombysepticus*, *B. thuringiensis* var. *kurstaki* + granulosis virus and *S. marcescens*. Overall, treatments with 2 g  $\cdot$  l<sup>-1</sup> or 2 ml  $\cdot$  l<sup>-1</sup> of the biocontrol agents were much less effective than the two higher concentrations (Fig. 2).

### Field trial

The five most effective BCAs from the laboratory experiment were evaluated in an *E. fordii* plantation damaged by *C. ombrodelta* (Fig. 3a, e). The site was highly homogeneous ( $P\% = 20.1$ – $21.6\%$  and  $DI = 0.22$ – $0.24$ ), and there was no significant difference ( $p = 0.990$ – $0.997$ ) in damage incidence and damage index between plots at the time treatments were applied (Table 3). Thirty days after trial establishment, there was no significant block effect, but there was a significant difference ( $p < 0.001$ ) in the damage incidence and damage index between treatments (Table 3). The *B. thuringiensis* var. *kurstaki* + granulosis virus and nuclear polyhedrosis virus treatments resulted in the least damage by *C. ombrodelta*, with damage incidence of 23.2% and 23.9%, and damage index of 0.83 and 0.85, respectively (Fig. 3b, c). The effectiveness of these two BCAs in reducing damage from the pest was over 70%. The other three agents (*B. bombysepticus*, *B. thuringiensis* var. *kurstaki*, and *S. marcescens*) were less effective at 57.2–64.9%, with damage incidence of 28.6–32.1% and damage index of 1.00–1.13 (Fig. 3d, f, g). By contrast, there was a significant increase in damage incidence ( $P\% = 77.4\%$ ) and damage index ( $DI = 2.91$ ) in the control treatment after 30 days (Fig. 3h).

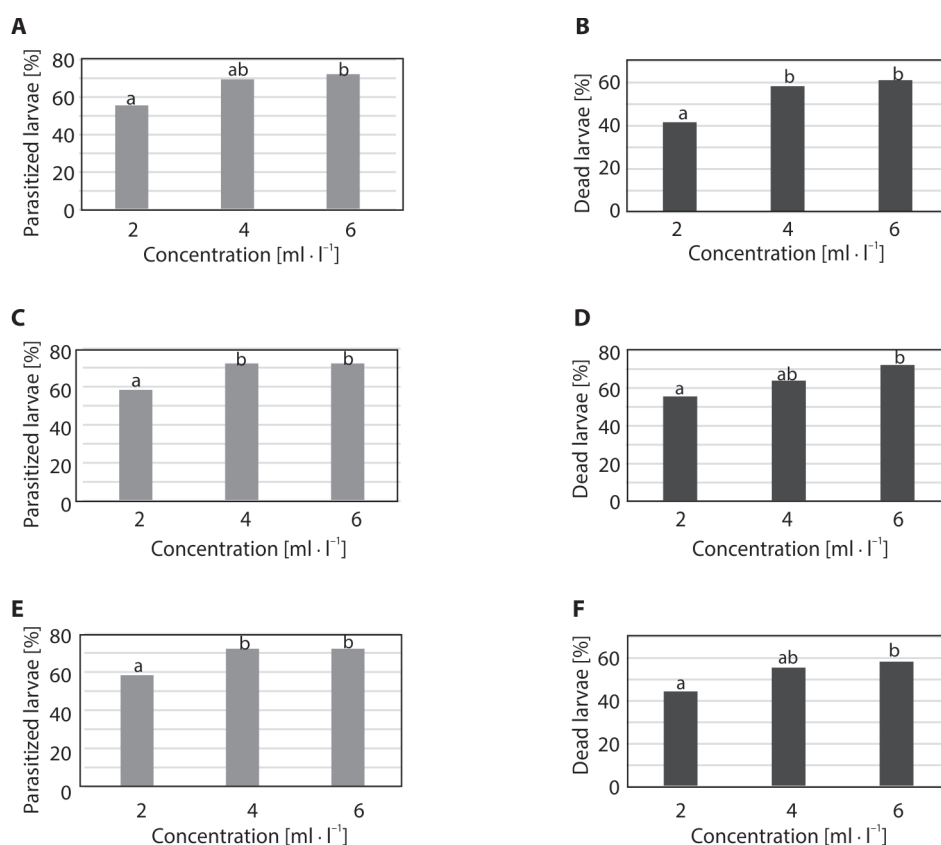
**Table 2.** Percentage of parasitized larvae with body discoloration and dead *Cryptophlebia ombrodelta* larvae at 7 days after spraying with biocontrol agents

| Biocontrol agent                                                | Parasitized larvae [%] | Dead larvae [%]    |
|-----------------------------------------------------------------|------------------------|--------------------|
| <i>Bacillus bombysepticus</i>                                   | 65.7 $\pm$ 10.00 b     | 53.7 $\pm$ 11.14 b |
| <i>B. thuringiensis</i> var. <i>kurstaki</i>                    | 67.6 $\pm$ 8.83 b      | 62.0 $\pm$ 11.07 a |
| <i>B. thuringiensis</i> var. <i>kurstaki</i> + granulosis virus | 70.4 $\pm$ 7.24 ab     | 63.9 $\pm$ 8.33 a  |
| <i>Beauveria bassiana</i>                                       | 48.2 $\pm$ 8.65 c      | 42.6 $\pm$ 9.28 c  |
| <i>B. bassiana</i> B2                                           | 41.7 $\pm$ 8.38 c      | 37.0 $\pm$ 8.45 c  |
| <i>Metarhizium anisopliae</i>                                   | 45.4 $\pm$ 7.43 c      | 39.8 $\pm$ 10.00 c |
| Nuclear polyhedrosis virus                                      | 76.9 $\pm$ 8.81 a      | 66.7 $\pm$ 9.48 a  |
| <i>Serratia marcescens</i>                                      | 67.6 $\pm$ 8.18 b      | 52.8 $\pm$ 8.15 b  |
| Sterilized water                                                | 0.0 d                  | 0.0 d              |
| p                                                               | <0.001                 | <0.001             |

Means within a column with the same letter are not different by Tukey's Multiple Range Test at  $p < 0.05$



**Fig. 1.** Effect of biocontrol agents on *Cryptophlebia ombrodelta* after 7 days of treatment in a laboratory experiment: A–E – external appearance of shoots before dissecting; F–J – dissected shoots showing appearance of larvae; F – larva has been dead for many days as the body is shriveled; G – larva is rotten with a dark color; h. larva is rotten with a light color; I – dry and dark parasitized larva; K – healthy larva; A, F – nuclear polyhedrosis virus; B, G – *Bacillus thuringiensis* var. *kurstaki* + granulosis virus; C, H – *Bacillus bombysepticus*; D, I – *Beauveria bassiana*; E, J – sterilized water



**Fig. 2.** Effect of the concentration of four biocontrol agents on the appearance of *Cryptophlebia ombrodelta* larvae after 7 days in a laboratory experiment: A, B – *Bacillus bombysepticus*; C – *Bacillus thuringiensis* var. *kurstaki*; D – *Bacillus thuringiensis* var. *kurstaki* + granulosis virus; E, F – *Serratia marcescens*. Means within a chart with the same letter are not different by Tukey's Multiple Range Test at  $p < 0.05$



**Fig. 3.** Effect of *Cryptophlebia ombrodelta* on the appearance of *Erythrophleum fordii* in the field: A, E – trees at the time the trial was established with new excrement, approximately 21% of attacked trees; B, C, D, F, G, H – 30 days after experiment; b. nuclear polyhedrosis virus; C – *Bacillus thuringiensis* var. *kurstaki* + granulosis virus; D – *Bacillus thuringiensis* var. *kurstaki*; F – *Bacillus bombysepticus*; G – *Serratia marcescens*; H – control, the main shoot has died. White arrows indicate damage symptoms caused by *C. ombrodelta*

**Table 3.** Comparison of biological treatments for their inhibition of *Cryptophlebia ombrodelta* damage to *Erythrophleum fordii* after 30 days in the field

| Biocontrol agent                                                | At treatment application |             | 30 days after first treatment application |               | Damage inhibition effect [%] |      |
|-----------------------------------------------------------------|--------------------------|-------------|-------------------------------------------|---------------|------------------------------|------|
|                                                                 | P%                       | DI          | P%                                        | DI            | P%                           | DI   |
| <i>Bacillus bombysepticus</i>                                   | 21.3 ± 1.2               | 0.23 ± 0.03 | 31.6 ± 3.1 a                              | 1.13 ± 0.09 a | 60.1                         | 60.7 |
| <i>B. thuringiensis</i> var. <i>kurstaki</i>                    | 21.2 ± 1.3               | 0.23 ± 0.02 | 28.6 ± 2.9 a                              | 1.00 ± 0.08 a | 63.7                         | 64.9 |
| <i>B. thuringiensis</i> var. <i>kurstaki</i> + granulosis virus | 20.8 ± 1.1               | 0.23 ± 0.04 | 23.2 ± 2.3 a                              | 0.83 ± 0.06 a | 70.0                         | 70.3 |
| Nuclear polyhedrosis virus                                      | 21.6 ± 1.4               | 0.24 ± 0.03 | 23.9 ± 2.6 a                              | 0.85 ± 0.07 a | 70.3                         | 71.6 |
| <i>Serratia marcescens</i>                                      | 20.1 ± 1.5               | 0.22 ± 0.05 | 32.1 ± 4.3 a                              | 1.13 ± 0.11 a | 57.2                         | 58.7 |
| Control                                                         | 20.8 ± 1.3               | 0.23 ± 0.03 | 77.4 ± 8.6 b                              | 2.91 ± 0.12 b | –                            | –    |
| <i>p</i>                                                        | 0.997                    | 0.990       | <0.001                                    | <0.001        |                              |      |

P% is damage incidence (%), DI is mean damage index. Means within a column with the same letter are not different by Tukey's Multiple Range Test at *p* < 0.05

Discussion

This is the first report on the use of biocontrol agents to protect *Erythrophleum fordii* trees from damage from *Cryptophlebia ombrodelta* (Lower, 1898) (Lepidoptera: Tortricidae). Previously, the Ba

Che Forest Company and Quang Ninh Department of Planting and Plant Protection have tried to control this pest with chemical insecticides, but it has not been very effective (data not published). This may be because *C. ombrodelta* has many generations through the year (Nguyen *et al.* 2023). Also,

many plantation owners avoid using chemical insecticides where trees are planted near community water sources.

In the laboratory trial, *Beauveria bassiana* and *Metarhizium anisopliae* resulted in <43% larval mortality. Therefore, they were not used for the field trial. These BCAs also showed low efficacy against *Hypsipyla robusta* (Moore, 1886) (Lepidoptera: Pyralidae) under greenhouse conditions (Chi *et al.* 2021a). However, in other studies they have been effective against many insect pests (Dara *et al.* 2019), including the control of *Aphis gossypii* (Glover, 1877) (Hemiptera: Aphididae) (Shaalan *et al.* 2021), *Frankliniella occidentalis* (Pergande, 1895) (Thysanoptera: Thripidae: Thripinae) (dos Santos Domingues *et al.* 2022), *Liriomyza sativae* (Blanchard, 1938) (Diptera: Agromyzidae) (Rajab *et al.* 2025), and in Vietnam against *Batocera lineolata* (Chevrolat, 1852) (Coleoptera: Cerambycidae) (Quang *et al.* 2022), *Neurozerra conferta* (Walker, 1856) (Lepidoptera: Cossidae) (Chi *et al.* 2022) and *Endocrita* spp. (Lepidoptera: Hepialidae) (Chi and Anh 2025).

Five BCAs (*Bacillus bombysepticus*, *B. thuringiensis* var. *kurstaki*, *B. thuringiensis* var. *kurstaki* + granulosis virus, nuclear polyhedrosis virus (NPV) and *Serratia marcescens*) caused 53–67% mortality in the laboratory trial and were further assessed in the field. *Bacillus thuringiensis* var. *kurstaki* + granulosis virus and NPV were the most effective BCAs in the field trial reducing *C. ombrodelta* borer damage to *E. fordii* trees by over 70%.

Previous studies where these BCAs have been beneficial include using *B. bombysepticus* to control *H. robusta* (Tra *et al.* 2022) and *Ceracris kiansu* (Tsai, 1929) (Orthoptera: Acrididae) (Chi *et al.* 2025); *B. thuringiensis* for *H. robusta* (Tra *et al.* 2022), and *H. grandella* (Zeller, 1848) (Lepidoptera: Pyralidae) (de Castro *et al.* 2018, 2019); *B. thuringiensis* var. *kurstaki*, *B. thuringiensis* var. *kurstaki* + granulosis virus for control of *Dasychira axutha* (Collenette, 1934) (Lepidoptera: Lymantriidae), *Dendrolimus punctatus* (Walker, 1855) (Lepidoptera: Lasiocampidae) and *H. robusta* (Chi *et al.* 2021a; Quang *et al.* 2021; Chi *et al.* 2023); granulosis virus against *Cydia pomonella* (Linnaeus, 1758) (Lepidoptera: Tortricidae) (Vail *et al.* 1991); and *Serratia marcescens* for control of *Episparis tortuosaalis* (Moore, 1867) (Lepidoptera: Erebididae) (Long *et al.* 2023). Furthermore, Moore (2002) found that a granulovirus was useful for the management of *Cryptophlebia leucotreta* (Meyrick, 1913) (Lepidoptera: Tortricidae), and this BCA is now being used commercially (Moore *et al.* 2015).

Overall, in the present study, NPV was the most effective BCA against *C. ombrodelta* in both the laboratory and field experiments. NPV has been widely used for control of insect pests with a high degree of host specificity and ability to produce epizootics

(Hamm 1993). This virus has been shown to be highly effective against *Heliothis* spp. (Lepidoptera: Noctuidae), *Pectinophora gossypiella* (Saunders, 1844) (Lepidoptera: Gelechiidae) (Bell and Kanavel 1977), *Malacosoma americanum* (Fabricius, 1793) (Lepidoptera: Lasiocampidae) (Progar *et al.* 2010), *Spodoptera litura* (Fabricius, 1775) (Lepidoptera: Noctuidae) (Ayyub *et al.* 2019) and *Helicoverpa armigera* (Hübner, 1808) (Lepidoptera: Noctuidae) (Nawaz *et al.* 2019). Marsberg *et al.* (2018) isolated a novel alphabaculovirus (containing NPV) from *C. peltastica* (Meyrick, 1921) (Lepidoptera: Tortricidae), and showed that it was an effective BCA against its host with LC50 of  $6.46 \times 10^3$  OBs · ml<sup>-1</sup> and LC90 of  $2.46 \times 10^5$  OBs · ml<sup>-1</sup> after 76–93 hours. This suggests an opportunity to find similar viruses with BCA potential in *C. ombrodelta* in Vietnam.

In this experiment, BCA dosages recommended by the manufacturers were used. In general, commercial BCAs in Vietnam are mainly supplied for the management of pests that damage agricultural and horticultural crops (Toan *et al.* 2024). Previously, Quang *et al.* (2022) and Chi and Anh (2025) identified limitations in control effectiveness when using these rates in forestry. Thus, further research is required in the forestry sector to ensure that optimum rates are applied for pest management in the future. As the price of BCAs formulated from bacteria, fungi and viruses are similar (Hai *et al.* 2023), the selection of which BCAs to apply should be based on efficacy and safety.

Vietnam has promoted the development of forest plantations with indigenous tree species (including *E. fordii*) from 2021 to 2030, with about 4,000–6,000 ha being established per year (Hai *et al.* 2023). Unless pest outbreaks are managed it is likely that *C. ombrodelta* will cause larger epidemics in Vietnam in the future. Recently, this polyphagous pest has been reported boring fruit of *Cassia fistula* (Tra *et al.* 2025) in areas where *E. fordii* is grown. Therefore, it is essential to investigate the management of this pest in greater detail. Factors that should be addressed include the timing and frequency of applications of BCAs across the seasons in plantations of different ages, the longevity and regeneration of BCAs in the field, and application of BCAs using drones. The trees used in the field trial were one year old, but the pest also damages older trees, and three years of integrated pest management may be required to prevent from damage to the boles. Lastly, finding, selecting, and screening natural predators also need to be considered to manage this pest.

## Conclusions

The shoot borer *Cryptophlebia ombrodelta* can be managed by the application of *B. thuringiensis* var. *kurstaki* + granulosis virus (4 g · l<sup>-1</sup>) or nuclear polyhedrosis

virus (4 ml · l<sup>-1</sup>). The percentage of parasitized and dead larvae at seven days after spraying in a laboratory were 70–77% and 64–67%, respectively. Application of these BCAs reduced borer damage symptoms in a one-year-old *Erythrophleum fordii* plantation by 70%.

## Acknowledgements

The authors wish to thank the Tam Dao Forest Company for their support in collecting samples and some data.

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