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Infestation and Evaluation Efficacy of Monitoring by Alcohol Trap and Application of Local *Beauveria bassiana* for Control of Coffee Berry Borer in the North-western Region of Vietnam

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ABSTRACT

Demand for higher food standards and concerns with heavy pesticide use led to a rapidly changing farming system in Vietnam. Overall, the percentage of berries damaged by CBB on the 1st and fourth flowering was higher than others. The highest infested berry percentage in the 1st flowering was 19.7, the second, third and fourth proportions infested berries were 9.21, 12.43 and 17.14%, respectively. The catched CBB adults/trap were highest in May and June, with an average of 7.5 to 101.4 CBBs per trap/ week. The highest proportion of berry infestation (5.9 to 7.3%) was recorded in October. The trials that control CBB using *Beauveria bassiana* showed a lower percentage of berry damaged by CBB (6.33) than the percentage of berry damaged by CBB on plot application of chemical insecticide (13.70). Based on these results, alcohol trap and *B. bassiana* should be implemented as a part of integrated pest management CBB to reduce the use of chemical insecticides towards sustainable food systems. Bottle traps using alcohol attractants will help farmers track CBB populations and monitor CBB adult flight to determine the best timing for insecticide (chemical, entomopathogenic fungi) spray. It was also concluded to assess CBB infestation and determine the efficacy of monitoring CBB by alcohol traps and the local *B. bassiana* for control of CBB.

Key words : Alcohol trap, coffee berry borer, *Beauveria bassiana*

INTRODUCTION

According to the International Coffee Organization report, the most significant coffee producing countries include Brazil, Vietnam, Colombia, Indonesia, India, etc. (Krishnan, 2017). Coffee berry borer (Ferrari) was reported as the most dangerous insect-pest of coffee (globally) and causing damage more than US$ 500 m every year. CBB damaged the yield and the quality of coffee products (Vega *et al.*, 2015). CBB makes holes to enter inside for damaging beans. The results are very clear and proven that CBB can act as a vector and carry the spread of the mycotoxin-producing fungal spores onto the coffee beans. The invasions, detection and responses in Indonesia, Brazil and Ethiopia (which are three crucial coffee-growing nations) were invaded early by CBB spread before the year 2007; Puerto Rico (Caribbean Sea/Atlantic Ocean) in the year 2010; Hawaii in the year 2017; and Papua New Guinea in 2020 (Melissa *et al.*, 2020).

It is hard to manage coffee berry borer (extremely hard), and the reason is that all of the cryptic life cycles are in the coffee berry (Aristizábal *et al.*, 2017). The integrated pest management (IPM) strategies for controlling the CBB approach were reported, including practices of culture control, focused on post-harvest sanitation/crop of berries infested, monitoring, biological control agents using chemical insecticides as well as cultural control (Vega *et al.*, 2015; Melissa *et al.*, 2020). Son La Province, located approximately 300 km westward from Hanoi and borders Laos to the south, is the largest province in the north-western region with a total area of 14,055 km^2 and a provincial population of 1,024 million. Thai ethnic minority people account for approximately 55%, more than half of the provincial population. The main crops are cassava, sugarcane and coffee beans. Although generally, the income of Vietnamese farmers is low, 87% of the workforce in Son La province is engaged in agriculture. The coffee berry borer had been recorded in Son La, causing coffee bean to be damaged and resulting in a

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10% coffee yield. Chemical pesticide has been used popularly for control CBB (Perfecto *et al.*, 2014). Chlorpyrifos and methidathion are the most efficient and widely used insecticides for control CBB (Suu *et al.*, 2021a). Recently, farming systems are rapidly changing in Vietnam due to a demand for higher food standards and concerns with heavy pesticide use. Methidathion and chlorpyrifos have been removed in Vietnam's agricultural production due to its high mammalian toxicity and environmental damage (Suu *et al.*, 2021b). So making much of high practicality and reasonability to practice IPM for production approach of safe as well as quality coffee was tried in this study. There are many nations that used Trapsto monitor CBB, and in some cases, to manage these populations by "Mass-trapping" to reduce females lead to reduce next generations. Worldwide, semiochemicals and entomopathogenic fungi have been extensively used in various cropping systems to reduce insecticides. Many factors affect trapping, entomopathogenic fungi efficiency, such as shade, spacing, wind direction, speed, climate, cultivar, plant age, etc. (Horgan, 2020). However, traps have not been studied on the slope fields in the north-western part of Vietnam. Monitoring of CBB infestation by alcohol traps and *Beauveria bassiana* for the control was focused in this report. Monitor the CBB adult flight to determine the best timing for insecticide (chemical, entomopathogenic fungi) spray. Cultural control is also recommended as the most crucial part of practicing IPM.

MATERIALS AND METHODS

Studies were conducted in coffee farms in the Chieng Ban village (21°14′20″N 103°55′57″E), Mai Son district, Son La province. The climate was sub-tropical, with a rainy season extending from April to October and a mean annual temperature of 21°C. The Cartimor variety was cultivated with high density (5000 trees/ha) coffee in the entire sunlight system (monoculture) and 1 ha coffee intercropped with fruit trees, long plum, the plants were 10 years old and about 1.7 m high.

CBB infestation rates were recorded once a week, beginning in the coffee season (April - May), continuing till the harvest (December-January). Thirty trees were randomly selected for assessment of each trial. Four branches in the mid-portion of each tree containing 30 - 50 developing green berries were chosen in the study. The tiny hole of entrance in the berry made by CBB adults was identified easily as symptom for counting infested and non-infested berries to find the proportion of berry infestation. One thousand berries growing in each of four flowering blooms were marked and counted for infested and non-infested berries throughout the coffee season.

The traps were formed using 2.0 litter transparent plastic bottles (initially of soft drink), and made a 13 x 18 cm small window over the bottom 9.0 cm. Two ml liquid detergent with 200 ml water was added at the bottom in the bottle trap. It was for preserving dead adult CBB. A 15 ml amber glass vial with a rubber cover (originally used as a medicine antibiotic powder container) was used as an alcohol dispenser. About 868 mg per day absolute ethanol and methanol (1:1) were used inside bottle. The vial was hung 20 cm above the trap bottom. Twelve m distance was kept between traps within a block of 30 m. The plot was put in a randomized design with three replicates. Twenty traps were arranged in every coffee farm. Weekly water with liquid detergent and alcohol was replaced. Water was removed with dead insects for counting. Vials were weighed to compute volatile release rates. Nuber CBB adults captured per trap and proportion berry infestation were recorded once a week.

The *B. bassiana* Bb5MCB1 was isolated from infected CBB samples collected from coffee plantations in the Son La province. Fungi were grown on PDA media at 27°C. The *B. bassiana* isolates were identified based on phenotypic and molecular data. The local *B. bassiana* Bb5MCB1 was inoculated on the cooked rice (autoclaved for 30 min at 120°C).

The experiment was arranged in tree coffee farms as three replicates. The treatments were (a) application of local *B. bassiana* as a part of an integrated pest management and (b) complete protection using chemical insecticide (15 g/l of cyhalothrin; 20 g/l of phoxim and 450 g/l of profenofos) three times in April, June and August. *B. bassiana* was sprayed on all parts of tree and ground three times (1st when there was mass movement of CBB as indicated by trap catch; 2nd in case berries were 60 days old and 3rd when berries were 120 days old). Proportional berry

infestations were recorded once a week. Captured CBB adults ratio (%) of infested berries was calculated. Pearson's correlation (r) was used for computing level of infestation. Generalized linear mixed models (GLMM) were used with binomial and poisson error distributions to compute proportion of berry infestation on plot application and complete protection using chemical insecticides.

RESULTS AND DISCUSSION

The rate of berries damaged by CBB on the 1st and fourth flowering was higher than others. The highest infested rate in the 1st flowering was 19.7%, the 2nd, third and fourth proportion infested berries was 9.21, 12.43 and 17.14%, respectively. On the other hand the survey results showed that the CBBs started to attach the berries 60 days after flowering (1st flowering); 88 days (2nd flowering); 89 days (3rd flowering) and 91 days (4th flowering). Percentage of berry infestation increased through time, and the pick was observed in September 2016 (Fig. 1).

Scientists observed that there were four flowering seasons in coffee to use bottle traps or alcohol traps to attract CBB (Fig. 2). Infestation of CBB fluctuation was also generally surveyed from April 2019 to January 2020. Scientists took into account 60 days as the period after first main flowering. The CBB started to attack berries on May 25th 2019. The infestation ratio increased over time and was highest (17.92%) on 2nd November 2019 (Fig. 3).

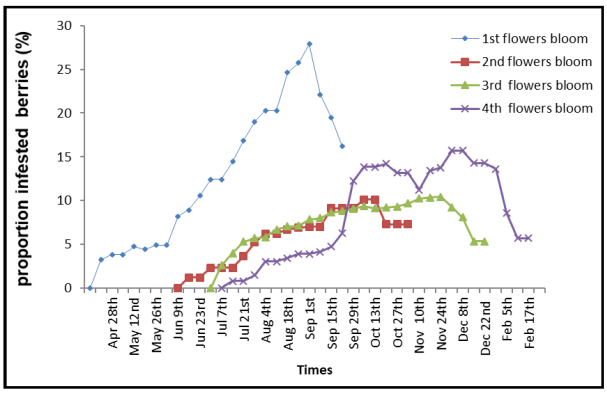

Fig. 1. Percentage of infestation of the CBB *Stephanoderes hampei* Ferrari on main coffee flowering during season 2016 and 2017 in Son La Province, Vietnam.

The activity of flight was recorded in a survey for CBB monitoring dispersing, *S. hampei* Ferrari (Coleoptera : Curculionidae : Scolytinae) with alcohol-baited bottle traps plus quantified ratio (%) of berry infested on four farming households of coffee growing in two locations in Son La province, Vietnam. The mean number of CBB adults was highest in

Fig. 2. The shape of berries in the 2016 and 2017 seasons in Sun La Province, Vietnam.

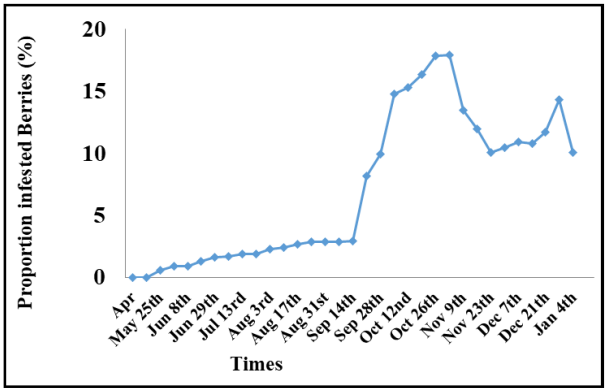

Fig. 3. Proportion infested berries fluctuation during the seasons of 2019 and 2020 in Son La Province, Vietnam.

May and June, with an average of 7.5 to 101.4 CBB for each trap per week. The average highest infested berry (5.9 to 7.3%) was recorded in October. Flight activity was highest during 2 to 3 months in the post-harvest season (April through May) when berries were only young on coffee plants. The proportion of infected CBB on two farms, where culture control (pick up of all cherry to fall to the ground or remain on trees; pruning) was practiced was better and lower than remaining on coffee farms (Fig. 4). A linear correlation (positively) between number of CBB adults caught, and ratio (%) of berry infestation was observed ($r^2 = 0.3035$; Fig. 5).

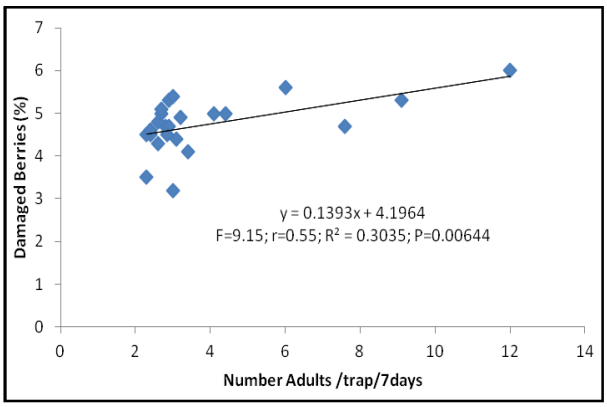

Fig. 5. The regression of the mean of infested coffee berries proportion and the number of CBBs captured.

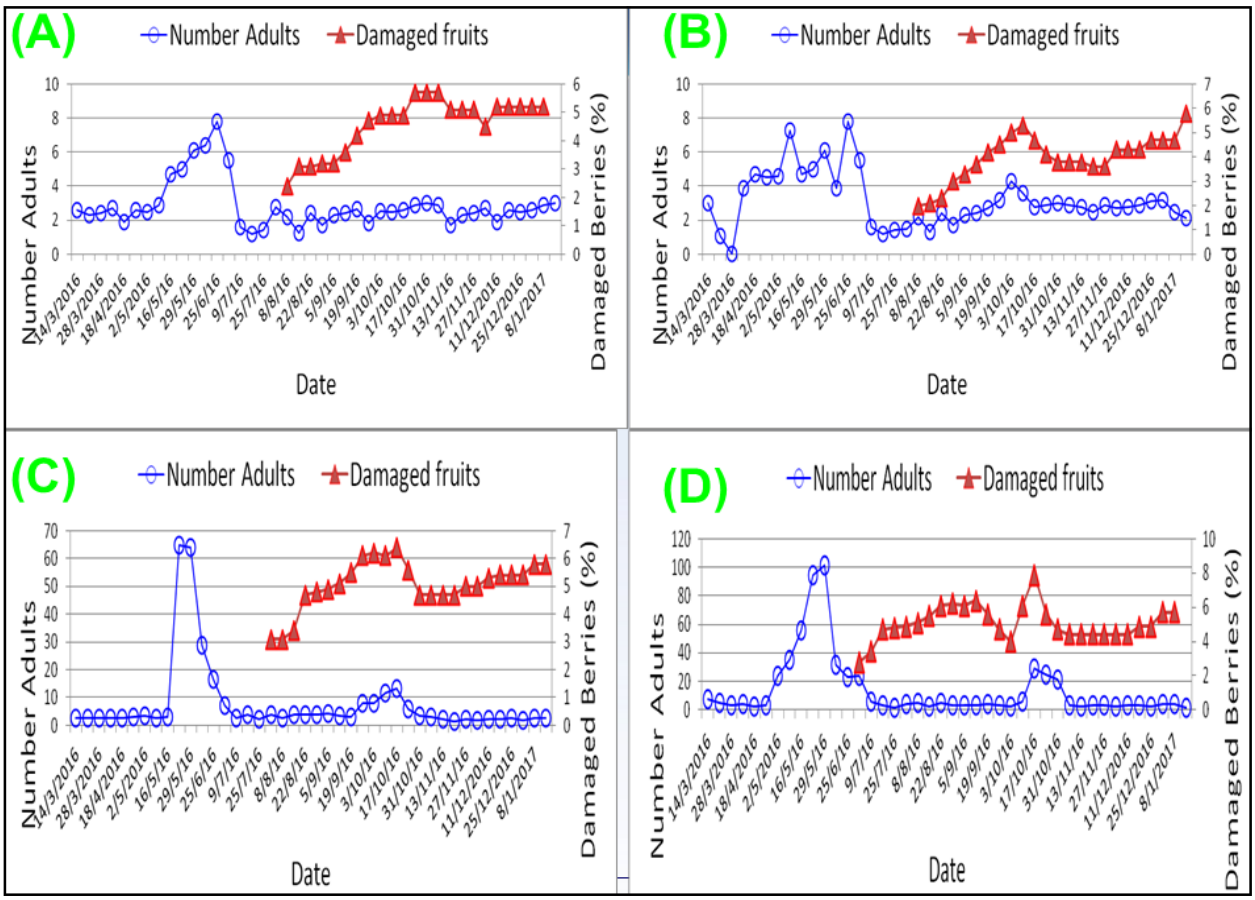

Fig. 4. The average number of adult coffee berry borers captured per trap per week and proportion infested berries fluctuations on four farms in coffee season of 2016-17 in Son La, Vietnam.

Fig. 6 shows the percentage of berry damaged by CBB which was different between every treatment. Scientists found 13.7% as a ratio of berry damaged by CBB, highest on chemical insecticide plot application which was 13.70%. Furthermore, the ratio of 6.33% was a lower percentage of berry damaged by CBB, in application of *B. bassiana*.

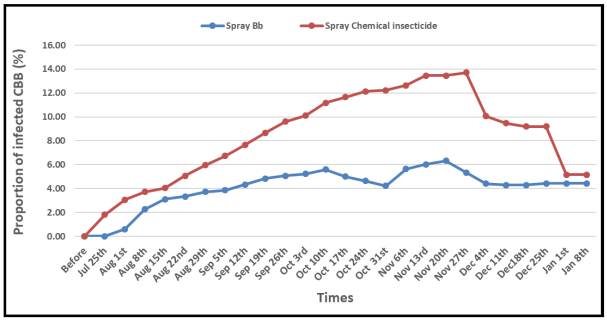

Fig. 6. The proportion of infested CBB (%) in plots application of local *Beauveria bassiana* and plots application of chemical insecticide.

Worldwide, semiochemicals and entomopathogenic fungi have been extensively used in various cropping systems to reduce insecticides. The research found out that *B. bassiana* significantly caused a decrease in insect population and the level (damaged) of CBB if attractants were used to monitor insect adult flight to determine the best timing for insecticide spray, the mass-trapping of pest (Fig. 7).

The result of infestation was similar to the previous studies. In Puerto Rico, the infestation in 2010 and 2011 was 4 to 26% in sun coffee. The proportion of berry damaged by CBB went up over time and highest was in November (Yobana *et al.*, 2015). It stated that the ratio (%) in Mexico in 1997 was 0.1-19% and 5-35% in 2008 (Rodríguez *et al.*, 2013).

The correlation between the number of adults catched (CBB), and ratio (%) of berry infestation showed the pest status in the field. It was used for monitoring CBB infestation. These results demonstrated the prediction level of CBB infestation in berries based on the number of CBBs in traps. The number of adult captures increased in consecutive samples. The ratio (%) of berry infestation, undoubtedly increased as well captures increased in consecutive samples. Correlation between CBBs captured, and the proportion of berry infestation was also observed for both large ($r^2 = 0.90$) and small ($r^2 = 0.68$) farms in the central coffee region of Colombia (Aristizábal *et al.*, 2017). The highest proportion of berry infestation was observed during January and February which was not observed in the present study ($r^2 = 0.3035$). It was due to climate differences. The Arabica variety was grown 100% in north-west Vietnam. The four central flowers usually bloom from March to April, depending on the climate, especially the rainy season. The number of CBB females was low in four farms between July and December, and it went up considerably between April and May. In contrast, the emergence of CBB from the dry berries remaining on plants or falling on the ground was experienced. Sixty days after flowering was the best time to start sampling, usually in May/June and Serrato-Diaz *et al.* (2020) suggested entomopathogenic fungus *B. bassiana* application to control CBB. In the coffee season, May till July was considered best

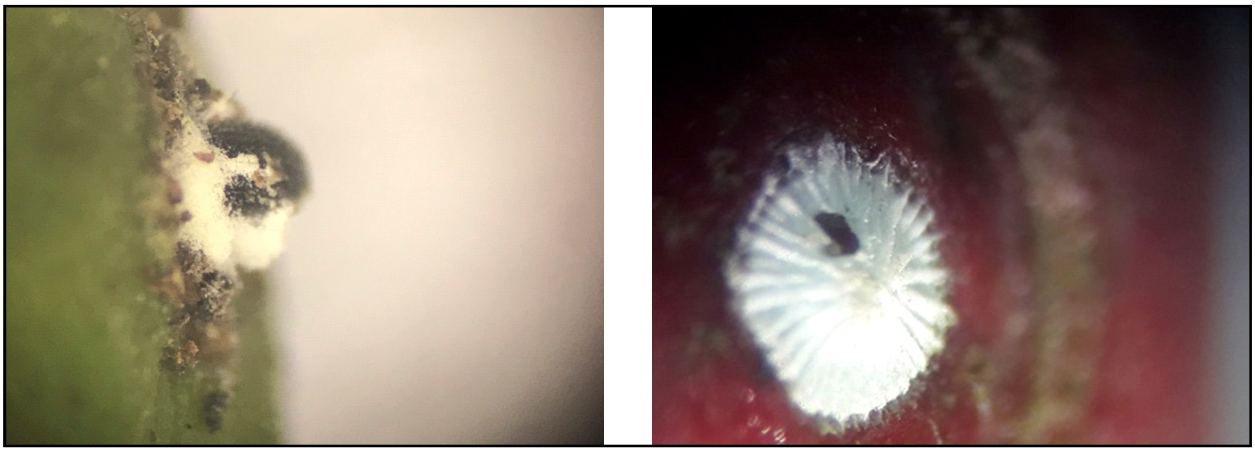

Fig. 7. Coffee berry borer was damaged by local *Beauveria bassiana*. Photo taken in Son La Province, Vietnam.

time or applying fungus. Hollingsworth *et al.* (2020) also recommended reducing CBB populations during the coffee season in case *B. bassiana* applications were alone or mixed with other insecticides. Further, application in the early season of coffee (April-July) compared to the second semester (August-November) was more effective.

CONCLUSION AND RECOMMENDATIONS

The percentage of berries damaged by CBB on the 1st and fourth flowering was higher than others. The catched CBB adults/trap were highest in May and June. The highest proportion of berry infestation was recorded in October. The trials to control CBB using *B. bassiana* showed the lower percentage of berry damaged by CBB than the percentage of berry damaged by CBB on plot application of chemical insecticide. Based on these results, alcohol trap and *B. bassiana* should be implemented as a part of intergreated pest managerment for CBB reducing than the use of chemical insecticides toward sustainability of food systerm. Bottle trap using alcohol attractants will help farmers to kill CBB females and monitor CBB adult flight to determine the best timing for insecticide (chemical, entomopathogenic fungi) spray. Among important crops in Vietnam, one has to mention the coffee crop as the main cultivated crop in the north-west area of the country to deal with the negative factor, or insect, coffee berry borer (CBB), which causes loss and damage to farmers.

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