



Available Online at EScience Press

Plant Protection

ISSN: 2617-1287 (Online), 2617-1279 (Print)
<http://esciencepress.net/journals/PP>

Research Article

EFFICACY OF A BIOETHANOL-METHANOL MIXTURE FOR TRAPPING COFFEE BERRY BORER IN ARABICA COFFEE PLANTATIONS IN INDONESIA

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ARTICLE INFO

Article history

Received: 11th September, 2024Revised: 28th October, 2024Accepted: 25th November, 2024

Keywords

Application attractant

Coffee berry borer

Population

Fruits damage

Arabica coffee

ABSTRACT

The coffee berry borer (CBB), a primary pest in Indonesia, causes significant damage to coffee fruits and beans, necessitating effective control measures. This study evaluated the efficacy of a synthetic bioethanol-methanol mixture trap for capturing beetles and mitigating their impact. The experiment was conducted in two plots: Plot A, with a *Leucaena leucocephala* shade density of 19.5%, and Plot B, with a shade density of 13.5%. The attractant mixture used in the traps was prepared in a 1:2 ratio of bioethanol to methanol. Attractant traps were hung on coffee trees at a height of 1.2 to 1.5 meters above the ground, and beetle populations were collected weekly over a nine-week period. Damage to coffee fruits and seeds was assessed in the first, fourth, and eighth weeks. The extent of seed damage was categorized into two groups: $\leq 50\%$ damage and $> 50\%$ damage. Plot A, with denser shade, exhibited a significantly higher beetle capture rate of 83.26%, compared to 16.74% in Plot B. Beetle population dynamics fluctuated, peaking during the sixth week of trapping and declining thereafter until the ninth week. The higher shade density and denser coffee canopy in Plot A were associated with an increased beetle population. The use of the bioethanol-methanol attractant traps significantly reduced seed damage in both damage categories ($\leq 50\%$ and $> 50\%$). The results demonstrate that the bioethanol-methanol mixture is an effective tool for monitoring and controlling CBB populations, thereby minimizing fruit and seed damage.

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INTRODUCTION

Indonesia is the fourth-largest coffee producer globally, following Brazil, Vietnam, and Colombia. Coffee is a crucial agricultural product in the economy of Indonesia, generating income for local communities, foreign exchange for the nation, and

providing employment opportunities. Arabica coffee, known for its low caffeine content and rich aroma, accounts for approximately 60% of the annual global commercial production. This variety is regarded as one of the highest quality and commands a relatively higher price compared to other types. However, the

coffee berry borer (CBB), *Hypothenemus hampei* (Coleoptera: Curculionidae: Scolytinae), is a major pest in all coffee-producing countries worldwide (de Souza et al., 2018, 2020). CBB infestations are a significant constraint on coffee production, often resulting in reduced quality. In Indonesia, coffee productivity remains relatively low at 825 kg/ha, falling short of its potential production capacity of 1,500-3,000 kg/ha (Campuzano-Duque et al., 2021). This invasive pest causes damage to coffee fruits ranging from 15% to 50%, and in some cases, up to 100%, including during storage (Wiryadiputra et al., 2009). Infested coffee fruits suffer damage to both the skin and beans, leading to a decline in quality, with poorly managed plantations experiencing quality reductions of over 80% (Silva et al., 2014). In North Sulawesi, CBB has caused fruit damage ranging from 65% to 100%, leading to a decrease in both the quality and quantity of coffee production. The adverse effects of this beetle pest reduce bean weight and alter both the taste and the type of coffee beverage (Vega et al., 2009). Defective coffee beans significantly impair their chemical composition, particularly in terms of caffeine levels and reducing sugars.

Pest infestations have significantly reduced coffee harvest yields in several countries, with losses reported at 80% in Uganda, 60% in Colombia, 58-85% in Jamaica, 90% in Tanzania, 50-90% in Malaysia, and 60% in Mexico (Vega, 2004). Globally, the coffee industry incurs annual losses exceeding USD 500 million due to damage caused by the CBB (Vega et al., 2009, 2015).

This pest uses coffee fruits and beans as a shelter, egg-laying site, food source, and breeding ground, where it completes its metamorphosis. Adult females colonize coffee fruits and lay eggs inside the beans. The larvae and emerging beetles consume the beans, significantly reducing both yield and quality, ultimately impacting the income of farmers worldwide.

Pesticide use has proven largely ineffective for CBB control because the pest reproduces continuously within ripe orange and red coffee fruits. Climatic factors also contribute to population abundance, while coffee trees over five years old and taller than a person pose logistical challenges for effective pesticide spraying.

Various control strategies have been employed, including cultural, biological, chemical, and post-harvest sanitation methods (Aristizabal et al., 2015). In

some coffee-producing countries, the insecticide endosulfan was once widely used to manage CBB. However, its use led to pest resistance and severe environmental pollution, resulting in its discontinuation.

Insect communication is facilitated by two types of attractant chemical compounds: kairomones and pheromones. Kairomones are attractants emitted by one species to lure individuals of another species, whereas pheromones are involved in intra-species attraction. The application of attractant compounds presents an effective and environmentally friendly pest control strategy for population suppression. This approach utilizes aromatic compounds capable of attracting females to a specific source. However, many Indonesian farmers remain unfamiliar with the use and application of attractant traps.

Bioethanol, a highly volatile chemical compound, plays a significant role in attracting CBB. In particular, mixtures of bioethanol and methanol have proven effective in attracting CBB populations in robusta coffee (*Coffea robusta*) plantations (Rimbing et al., 2021a). Synthetic alcohol-based attractants, such as ethanol-methanol formulations, are considered effective tools for reducing pest populations in coffee plantations (de Souza et al., 2018, 2020; Ruiz-Díaz and Rodrigues, 2021). Transparent traps baited with a methanol-ethanol mixture (3:1) yielded the best results, capturing 14.3 ± 5.4 adults per trap per week, outperforming traps baited with ethanol and 40 g of ripe robusta coffee berries (Carvalho et al., 2023).

The present study aimed to evaluate the effectiveness of a synthetic bioethanol-methanol attractant formula in capturing beetle populations and to assess its impact on damage to coffee fruits and beans following attractant application.

MATERIALS AND METHODS

The study was conducted on *Coffea arabica* (Arabica coffee) plants in Minahasa, North Sulawesi Province, Indonesia, located at coordinates 1°01.18748'N, 124°0.81013'E. The coffee plants were grown in a monoculture system due to their ability to thrive even without the shade provided by trees such as lamtoro (*Leucaena leucocephala*). The study site is situated at an altitude of approximately 675 meters above sea level, providing ecological conditions favorable for the development and growth of CBB. The research was

conducted on fruit-bearing coffee plants that had been in production for approximately 15 years, although the specific variety cultivated by the farmers was not documented.

Sampling method

The experiment utilized a plant-based ethanol-methanol synthetic attractant to lure CBB and non-target insects. The bioethanol-methanol mixture, previously shown to attract beetle populations on *C. canephora* (robusta coffee) plants, was tested for its efficacy in this study. The attractant mixture consisted of 50% bioethanol and 90% methanol in a 1:2 ratio (1 part bioethanol to 2 parts methanol).

The research area covered 3,325 m², divided into two plots: Plot A and Plot B. Plot A spanned 1,700 m² with a planting distance of 2 × 1.5 m (566 coffee trees), while Plot B covered 1,655 m² with a planting distance of 2 × 2.0 m (385 coffee trees). The two plots were separated by a distance of 25.5 meters. The density of shade trees (*L. leucocephala*) was 19.75% in Plot A and 13.5% in Plot B.

Trap bottles with a capacity of 1,500 ml were used, featuring 5 × 5 cm holes on opposite sides to allow the entry of CBB and non-target insects. Each trap bottle was filled with 200 ml of water and 2 ml of liquid detergent to immobilize and capture insects. Smaller bottles (25 ml) containing the attractant mixture (12.5 ml) of bioethanol-methanol were suspended inside the larger trap bottles, following the method described by Rimbing et al. (2021a). The traps were hung on coffee trees at a height of 1.2 to 1.5 meters above the ground.

The bioethanol used in this study, branded locally as "Ethanol Cap Tikus", is a unique product of North Sulawesi, distilled by farmers from the sap of the sugar palm (*Arenga pinnata* Merr.).

Insect collection

Attractant traps, used as bait for beetles, are placed within the rows of coffee plants. A total of four traps are positioned in each row at 10-meter intervals, resulting in eight traps for two coffee plant plots. The trapped beetle population is observed weekly over nine consecutive weeks, spanning 0 to 63 days. During these observations, the traps are replenished with a bioethanol-methanol mixture and are checked weekly at 10:00 AM. The contents of the traps were collected and transported to the laboratory, where the captured insects were counted. Furthermore, 50 randomly

selected, infested coffee fruits from each plot are inspected to evaluate CBB population.

Coffee fruit damage

The percentage of damaged coffee fruits was assessed both before and after the placement of attractant traps. This evaluation involved collecting coffee fruit samples from trees, including dark green, orange, and red fruits. At each location where an attractant trap was placed, three coffee trees with abundant fruit were selected. Fruits were collected to measure the extent of damage based on the four cardinal directions (north, east, south, and west). At each point, 150-200 coffee fruits from three trees were collected. The collected fruits were dissected to examine the disc region bored by beetles, distinguishing between damaged and undamaged fruits. The level of damage to coffee fruits or beans was calculated for Plot A and Plot B and classified into two categories: ≤ 50% damage and > 50% damage (Figure 1). The following formula was used to calculate the damage percentage:

$$Ps = \frac{A}{B} \times 100$$

$$Ps = A/B \times 100 \%$$

Ps = Percentage of damage

A = Number of infested fruits/seeds

B = Number of healthy coffee fruits.

This methodology provides a comprehensive assessment of beetle population dynamics and their impact on coffee fruit damage.

Insect identification

Non-target insects preserved in 70% synthetic ethanol were sorted based on their taxonomic orders and identified using morphological characteristics with the aid of insect identification keys (Borrer et al., 1996; Kerruish and Unger, 2010; Wedad et al., 2019). Observations and identifications were conducted under a binocular microscope, allowing classification up to the family and genus levels.

Statistical analysis

A paired sample t-test was conducted to assess differences in insect populations and coffee fruit damage between paired samples. This approach, commonly applied in field research, evaluates whether there is a significant difference in the means of two related groups. A significance level of 0.05 was used as the threshold for determining statistical differences between treatments. All statistical analyses were performed using SPSS software (version 22).



Figure 1. Criteria for coffee beans damaged by coffee berry borers.

RESULTS AND DISCUSSION

The attractant trap not only attracted CBB but also captured various non-target insects. These non-target insects belonged to six orders and 20 families. Among them, members of the family Muscidae acted as plant pests and decomposers of organic matter. The genus *Carpophilus* was identified as a carrier of plant pathogens, while Staphylinidae included predatory insects that also contributed to the decomposition of organic matter. *Bactrocera* species, significant agricultural pests, and members of Formicidae, which function as both plant pests and predators, were also captured. These insects were the most dominant based on the attractant trap captures (Figure 2).

The use of bioethanol-methanol attractants demonstrated potential for controlling key pests of chili plants, particularly *Bactrocera* spp. and *Carpophilus* spp., which are known carriers of plant pathogens in Indonesian plantations. For example, *Carpophilus* spp. are vectors of *Phytophthora palmivora* Butl., a pathogen responsible for black rot in cocoa fruit (Rimbing et al., 2021b).

The population of CBB was not observed in green coffee fruits but was found in orange and red fruits. The green coffee fruits had soft endosperm and were penetrated for feeding prior to abandonment. A drill hole, approximately 1 mm in diameter, located at the discus or tip of a coffee fruit, significantly alters its characteristics. When the fruit is dissected, the seed becomes visible through the drill

hole. The endosperm of the coffee fruit hardens, providing a suitable environment for CBB reproduction. The life

stages of CBB within coffee fruits include eggs, larvae, pupae, and adult beetles (imago) (Figure 3).

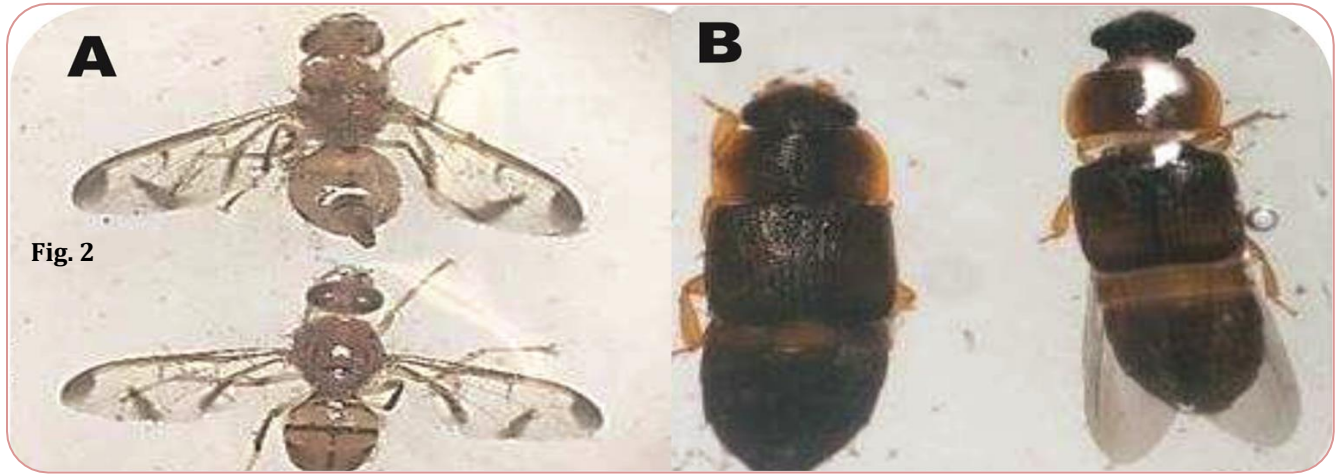


Figure 2. Non-target insects caught in bioethanol-methanol attractant mixture traps. A: *Bactrocera* sp, B: *Carpophilus* sp.

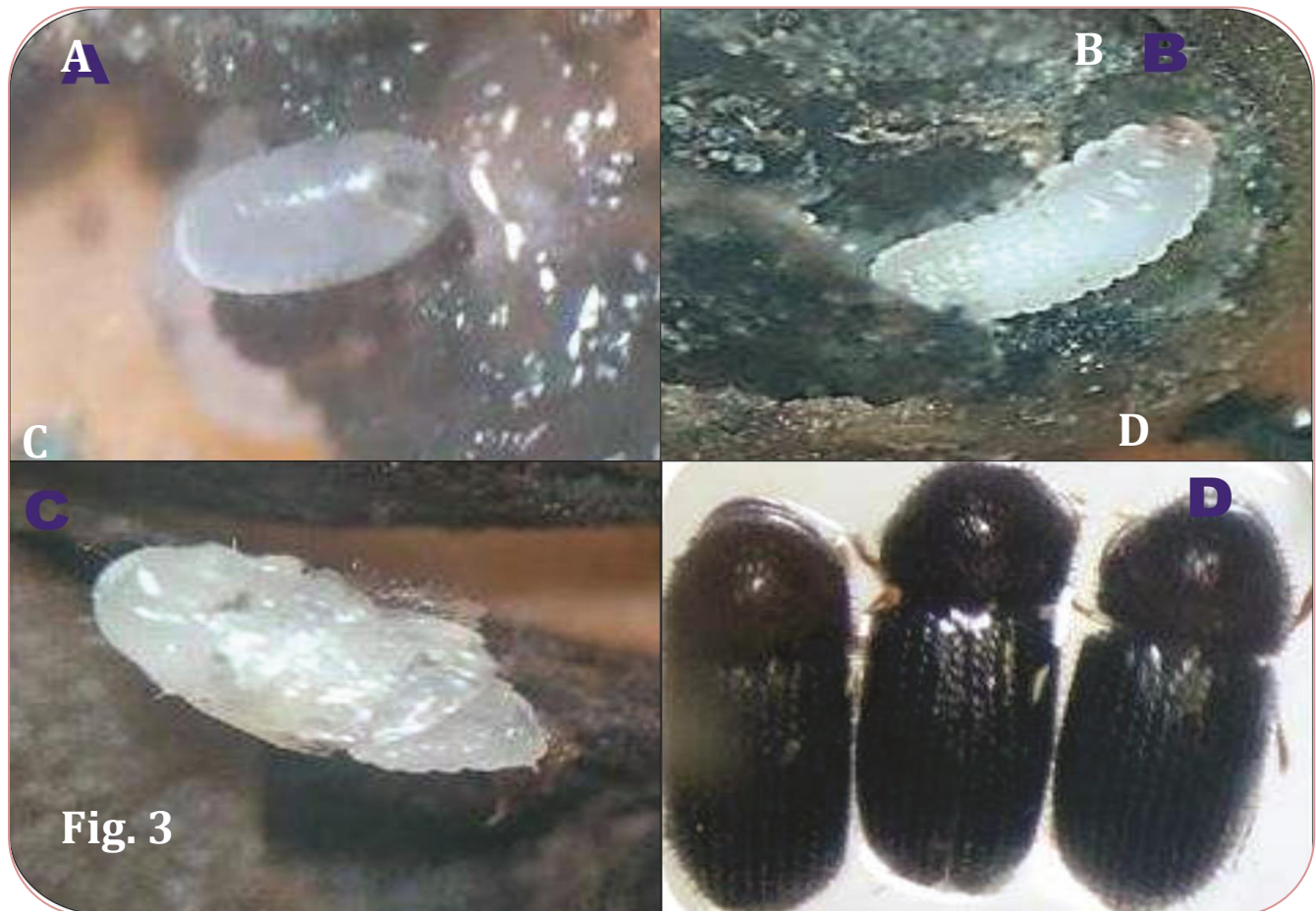


Figure 3. Stadium coffee berry borer. A: Egg; B: Larva; C: Pupa; D: Femele beetle.

The trapped CBB population was predominantly female, as males were not effectively captured. Male beetles, typically found inside coffee fruits, were unable to fly due to underdeveloped wings. The number of trapped individuals varied significantly, with plot A exhibiting a higher population compared to plot B. The maximum population captured per trap was 7.90% in plot A and 1.78% in plot B.

The trap mechanism involved insects entering a bottle trap and colliding with its inner walls, subsequently falling into a detergent solution at the bottom. This incapacitated their ability to fly, leading to their death. Traps baited with vegetable bioethanol and synthetic methanol attracted significantly higher CBB populations compared to traps using synthetic ethanol-methanol mixtures. Beetles are unlikely to develop resistance to bioethanol and methanol unless repellent

compounds are present.

Previous studies reported that synthetic attractant traps captured an average of 27 individuals per week (Rostaman and Prakoso, 2020). However, Sinaga et al. (2015) found a lower capture rate, ranging from 1.1 to 6.2 individuals per week. Variation in the captured insect population was observed, with moths often becoming dominant, thereby reducing the number of CBB captures. When moth populations exceeded two individuals per trap, captures were notably lower, particularly in plot B.

The combination of ethanol-methanol attractants with frontalin exhibited a repellent effect, deterring 77% of the CBB population (Njihia et al., 2014). The bioethanol-methanol mixture is hypothesized to contain chemical compounds with repellent properties, albeit in relatively low concentrations.

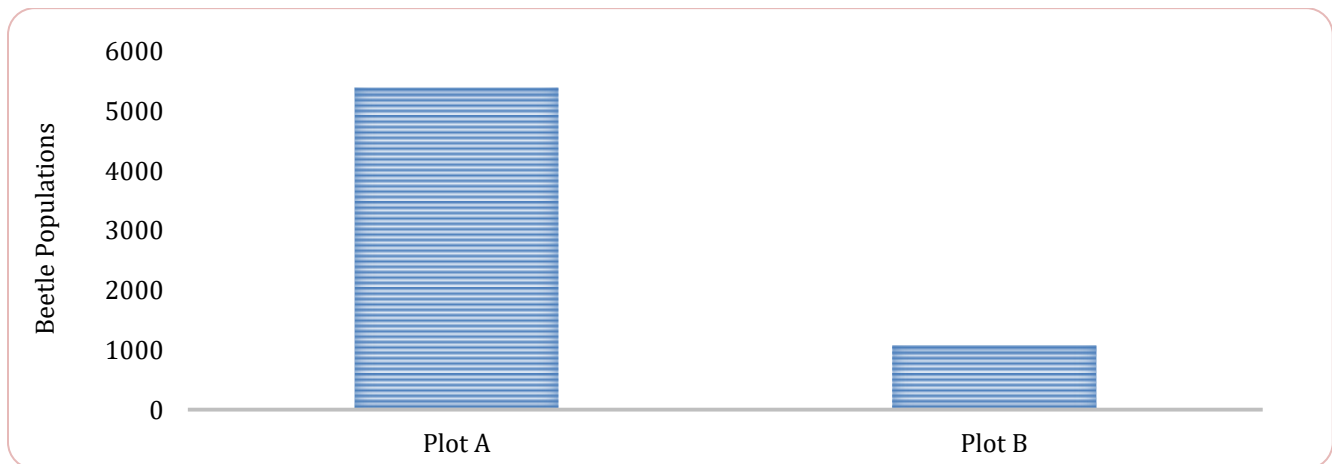


Figure 4. Total infestation by female coffee berry borer beetles over nine weeks of observation.

According to the paired T-test analysis, there was a significant difference between plot A and plot B, with a p-value of 0.003. The population in plot B was relatively low, as evidenced by the population captured over nine weeks, which amounted to 16.74%, while plot A accounted for 83.26% (Figure 4). Based on the examination of 50 coffee beans, the highest population was recorded in plot A. The total population captured by the bioethanol-methanol attractant trap over five weeks was 91.10% for robusta coffee, while the remaining insects were non-target species (Rimbing et al., 2021a). This high infestation in plot A was related to the density of *L. leucocephala* shade plants compared to plot B. The lower density of shade plants in plot B contributed to the

reduced population abundance. Coffee plants with shade densities above 40% were associated with a higher CBB population (Marino et al., 2016; Oliva et al., 2023). The infestation was also closely linked to environmental temperature, which affects insect physiology, abundance, and distribution. These temperature variations in coffee plants were closely associated with the density of shade plants. Consequently, measuring the density of shade plants provides valuable insight into determining coffee bean resistance and susceptibility.

In this study, the captured population was relatively high due to the monoculture planting of coffee at the experimental site. In monoculture systems, the CBB population was higher compared to polyculture systems,

such as coffee-orange intercropping, where the infestation level was reduced (Efrata et al., 2023). Generally, monoculture planting systems exhibited lower insect diversity compared to polyculture systems, which include predators and parasitoids. The population of ants, *Solenopsis* sp. and *Crematogaster* sp., which act as CBB predators (Constantino-Chuaire et al., 2022), was insufficient in the coffee bean boring holes, likely due to the monoculture system in this experiment.

In plot A, coffee trees were planted at a spacing of 2×1.5 m and were pruned, resulting in the highest beetle population. In plot B, the trees were planted at a spacing of 2×2.0 m. It appeared that the closer planting distance led to a higher beetle population. The Catimor variety experienced more intense attacks by CBB compared to the Caturra variety, suggesting that a denser coffee planting configuration enhanced the susceptibility to CBB (Oliva et al., 2023). In plot A, the canopy was dense with leaves and branches, leading to a compact arrangement of coffee plants. This dense canopy, with its thick foliage, blocked a significant amount of sunlight, causing a decrease in air temperature and an increase in humidity. These conditions resulted in a considerable increase in the beetle population. Variations in shade plant density and canopy growth affect the microclimate. Since some insects were poikilothermic and cannot regulate their body temperature, they were more effective at surviving under higher environmental temperatures. The beetle population was found to be low at temperatures below 20°C, but it increased as temperatures rose above 25°C

(Baker et al., 1992). The environmental conditions in plot A were conducive to population growth due to the close plant spacing, dense canopy, and increased shade.

Wind plays a critical role in the diffusion of attractants, facilitating their evaporation and allowing these substances to reach the sensory organs of the beetles. During the research period, the wind came from the east and south directions, with moderate strength, which positively impacted the beetles. A wind speed of approximately 2.5 m/s was positively correlated with population growth, although this relationship became negative at higher wind speeds (Johnson and Manoukis, 2021). The wind speed around the coffee plants was predicted to be less than 2.5 m/s, which resulted in the maximum capture of the population.

Population fluctuations were used to develop action thresholds and forecasting models for determining control measures. The female population was initially low during the first week, increasing from the second to the ninth week, with fluctuations in development over time (Figure 5). The female population fluctuated in both Arabica and Robusta coffee plants (Botelho et al., 2021; Rimbing et al., 2021a; Oliva et al., 2023), although the peak population occurred in the sixth week, gradually decreasing from the seventh to the ninth week. A significant population increase was observed in the sixth week in plot A (333 individuals, 24.70%) and plot B (53.75 individuals, 17.25%), followed by a decline from the seventh to the ninth week, with population proportions of 5.61% and 7.01%, respectively.

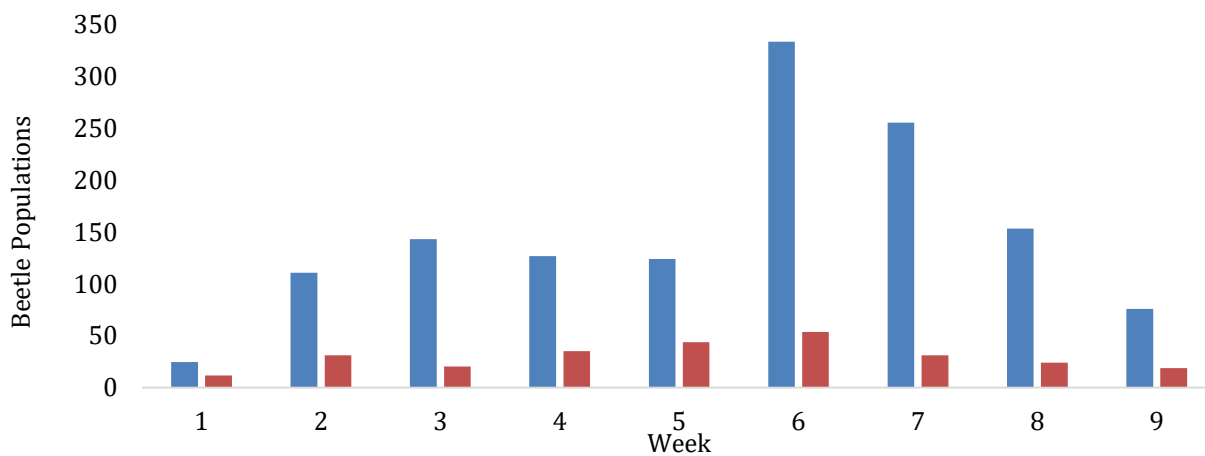


Figure 5. Population dynamics of coffee berry borer captured using attractant traps from week 1 to week 9 in plot A and plot B.

The study found that air temperature, influenced by the rainy season, had a significant effect on population growth, although it did not substantially impact overall growth. The Agricultural Extension Center recorded 226.8 mm of rainfall over two months, with 27 rainy days. However, population growth was not positively affected by the amount of rainfall or the number of rainy days. Temperature and relative humidity showed minimal variation throughout the year and did not significantly influence population dynamics (Botelho et al., 2021). Heavy rainfall (> 100 mm) hindered the flight of female beetles (Johnson and Manoukis, 2021). The number of rainy days could either positively or negatively affect the population, with a decrease in population observed during rainfall between 47.4 and 58.5 mm per day (Rimbing et al., 2021a). Furthermore, rainfall influenced the evaporation of attractants and beetle dispersion. Higher rainfall values slowed the volatilization of attractants and reduced the flight activity of Scolytinae beetles (Sanguansub et al., 2020). Rainfall also affected air temperature, causing it to decrease with higher rainfall and increase with lower rainfall. An increase in temperature promoted the evaporation of attractants, making it easier for the beetles to detect them. The peak population observed in the sixth week was not influenced by climatic factors but rather by the beetles' life cycle. In the second and third weeks of May, beetles began laying eggs. Over the course of a month, the larvae matured, reaching the peak population by the sixth week at the end of June. The captured beetles in the sixth week still displayed a light brown color, indicating that they had recently emerged from pupae and were seeking food. The development of CBB from egg to adult beetle typically took about 30-38 days (Erfandri et al., 2019; Hamilton et al., 2019). Although observations were limited to a small

scale over two months, the peak population occurred at the end of June, with the lowest recorded at the end of May in Minahasa. The synthetic attractant traps for beetle capture showed low effectiveness in March and July, with an increase in beetle capture observed in August, coinciding with the appearance of dried fruits on the trees and the ground (Pereira et al., 2012). The phenology of coffee plant growth also influenced the captured population, causing fluctuations during the ripening process. Bean damage negatively affected chemical compounds such as caffeine and reducing sugars. The size of the holes in the beans was a major factor in chemical quality damage, which, in turn, impacted the taste of the coffee beans. The seed damage caused by CBB (Figure 1) was categorized into two categories: ≤ 50% and > 50%. The intensity of seed damage by CBB before the installation of traps was notably high, as shown in Table 1, indicating the need for control measures. The high attack rate was attributed to the lack of control strategies, as no effective control technologies had been identified at the research site. According to Vega et al. (2012), control measures should be implemented when the attack rate exceeds 10%. An attack rate of 20% can result in a production loss of approximately 10%. After the installation of bioethanol-methanol attractants, from the first to the eighth week, there was a significant reduction in coffee bean damage, 83.30% in plot B and 49.12% in plot A, where the damage was ≤ 50%. According to Fernandes et al. (2014), effective attractant traps can reduce seed damage by up to 57%. This reduction in coffee bean damage contributed to an increase in the number of undamaged beans. The decrease in damage was most noticeable in the fourth week of May and the eighth week of June.

Table 1. Percentage of coffee fruit damage before and after trap installation

Location	Seed damage % (week)					
	First		Fourth		Eighth	
	≤ 50%	> 50%	≤ 50%	> 50%	≤ 50%	> 50%
Plot A	47,01 ± 2,43	18,63±3,27	41,72±8,76	21.12±7.94	23,92±9,19	8,08±2,75
Plot B	53,72 ± 6,87	12,67±6,31	15,83± 3,81	2,58±2,01	8,97 ± 2,56	4,37±1,32

The percentage of seed damage less than or equal to 50% is relatively higher than that of seed damage greater than 50%. CBB primarily causes seed damage in the ≤50% category, resulting in seeds turning black. Beetles and larvae feed on two seeds in a coffee fruit or

only consume seeds when the damage exceeds 50%. It is important to note that initial observations showed seed damage above 50%. However, after applying synthetic bioethanol-methanol traps, there was a significant reduction in seed damage during the fourth and eighth

observations. Seed damage is commonly observed in orange and red coffee fruits and serves as a breeding ground for CBB. In contrast, seed damage $\leq 50\%$ is rarely associated with breeding grounds, as this damage generally occurs in green coffee fruits. Most green coffee fruits exhibit seed damage ranging from 15% to 25%, falling within the $\leq 50\%$ category. According to the statistical analysis of the T-test from the first week of observation, no significant difference was found between seed damage $\leq 50\%$ and $> 50\%$ in plots A and B.

The significant difference began to emerge in the fourth and eighth weeks, with a p-value of 0.01 (Table 2). CBB assesses seed damage in coffee beans based on the defect value, which is a key factor affecting the quality of brewed coffee. Controlling beetles, as Novita et al. (2010) suggest, can reduce the defect value caused by physical damage. The application of synthetic bioethanol-methanol attractants to coffee plants has proven effective in capturing female beetle populations, thereby reducing coffee bean damage.

Table 2. Statistical analysis based on coffee fruits damage criteria from the first to the eighth week

Variable	T-test			p- value		
	First	Fourth	Eighth	First	Fourth	Eighth
$\leq 50\%$	2.114	9.955	4.024	0.125 ^{ns}	0.002*	0.028*
$> 50\%$	1.467	5.498	1.667	0.239 ^{ns}	0.012*	0.194 ^{ns}

Note : ^{ns} Not Significant, *Significant.

The determination of defective coffee beans involves identifying black and broken beans, which are often caused by CBB infestations (Aklimawati, 2014). When more than 50% of the coffee beans are affected by CBB, approximately one out of every two beans will turn black. Beans that are black, brown, or have holes significantly impact the taste, and when over 50% of the beans are damaged, they become uncompetitive in the market. To assess the quality of the coffee, the beans are soaked in water. If around 85% of the beans float, they are considered of good quality; otherwise, they are deemed defective. Once the beans are peeled, it becomes clear if more than 50% are damaged. It is essential to ensure that ground coffee is free from defective beans. Controlling beetles using bioethanol-methanol attractants is crucial to prevent damage to coffee beans.

CONCLUSION

The combination of bioethanol attractant traps with synthetic methanol effectively captures beetle populations, significantly reducing coffee bean damage. The presence of dense shade plants and coffee plant canopies influences beetle population growth, which in turn can affect the sensitivity and resistance of coffee beans to the coffee berry borer (CBB). Beetle populations caught over a period of nine weeks fluctuated, with the peak occurring in the sixth week. This peak is related to the CBB life cycle. Based on the results, the bioethanol-methanol mixture trap proves to be an effective tool for

capturing beetles, making it useful for both monitoring and controlling beetle populations. To suppress future beetle populations, bioethanol-methanol attractants can be applied, potentially in combination with other control methods.

ACKNOWLEDGMENTS

We authors acknowledge the support financially from University Sam Ratulangi, Indonesia.

AUTHORS' CONTRIBUTIONS

JR and JP designed the study, formulated the experiments, and executed them; FR and RE collected and organized the data, analyzed the results, and wrote the manuscript; SL and DK assisted in writing the manuscript and proofreading the paper.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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