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Assessing the Efficacy of Solid Lure Plugs and Insecticide Dispensers in Capturing Dacine Fruit Flies and Non-target Insects

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ABSTRACT

This study evaluated the effectiveness of solid male lures, specifically methyl eugenol (ME), cuelure (C-L), and tri-med lure (TML) combined with insecticides, for trapping fruit flies in three different green zones in Bangladesh. The study also investigated the non-target insect attraction to traps using these lures. The traps were set up across nine locations in three distinct experimental areas: i. Atomic Energy Research Establishment (AERE) colony, ii. AERE office campus, and iii. Jahangirnagar University (JU) campus, covering agricultural fields, backyard gardens, and mixed plantations. Data were collected weekly over 18 weeks from May to September 2015. The captured fruit fly species, including *Zeugodacus cucurbitae* (Coq), *Zeugodacus tau* (Walker), *Bactrocera dorsalis* (Hendel), and *Bactrocera zonata* (Saunders) were monitored, and non-target insect captures were recorded. The highest capture rate of dacine fruit flies was at the JU campus, with 98.41% of *B. dorsalis* (538.05 ± 62.28 fly/trap/week (FTW)) being captured using ME. C-L attracted higher numbers of *Z. cucurbitae*, while *Z. tau* was most attracted to C-L at both the AERE office campus and the JU campus. No *Bactrocera* species were attracted to TML. The majority of non-targets were saprophagous Diptera, such as *Drosophila* and Milichiidae, along with Hymenoptera (black ants), especially in traps baited with C-L and ME. The study showed that the lure responses were species-specific, with no adverse effects on beneficial non-target insects. The combination of DDVP strips and solid lures proved to be an effective method for mass-trapping dacine fruit flies.

Keywords: Insecticide strips, Solid lures, Non-target insects, Dacine, Capture, Tephritid fruit fly

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Introduction

Dacini fruit flies, part of the Tephritidae family (Diptera: Tephritidae), are primarily florivorous or frugivorous, with around 10% of the 932 recognized species being harmful pests of various fruits and vegetables. This genus includes several highly invasive and polyphagous species, such as the melon fly, the pumpkin fruit fly, *Bactrocera dorsalis*, also known as the oriental fruit fly, and the peach fruit fly among others. The genus *Ceratitis*, which has around 65 species found in tropical regions, also includes many pest species, notably the Mediterranean fruit fly, which has spread globally. This pest can be trapped using food-based bait in Hawaii, along with traps containing varying amounts of trimedlure (TML) [1-5].

Economically, fruit fly species from these genera cause considerable damage to crops, enforce quarantine restrictions on affected areas, necessitate postharvest treatments for commercial fruits before export, and serve as breeding grounds for the spread of these pests to other regions [6, 7]. The spread of these pests has been exacerbated by increased global trade and human travel. These pests have been controlled or even eradicated in some areas through the area-wide deployment of male lures, and male annihilation techniques (MAT) have also been used for their suppression [8-12].

Among the most commonly used male lures for detecting Tephritids are TML (tert-butyl 4- and 5-chloro-cis- and trans-2-methylcyclohexane-1-carboxylate), raspberry ketone (RK), Cue-lure (C-L), and Methyl eugenol (ME). These lures are highly effective at attracting male fruit flies of specific species, such as *Bactrocera* fruit flies being drawn to C-L/RK or ME, while TML attracts various male *Ceratitis* species. ME is a natural plant product found in over 200 plant species in tropical regions, while C-L, though not naturally occurring, breaks down rapidly into RK, a potent lure for *Zeugodacus cucurbitae*. Recent studies have shown that C-L remains intact in the atmosphere long enough to act as a lure [13-16]. Additionally, C-L has been identified in daciniphilous flowers such as *Bulbophyllum hortorum* and *Passiflora maliformis*. Furthermore, a novel fluorinated derivative of raspberry ketone, raspberry ketone trifluoroacetate (RKTA), has been found to attract more *Bactrocera tryoni* than C-L or melolure. Of the 54 Dacini species that are pests, 16 are attracted to ME, while 26 respond to C-L/RK [17-22].

In fruit fly detection and control efforts, various trap types are commonly used, which are baited with male lures and often combined with toxicants in liquid form. Some of the most widely used traps for detecting fruit flies with lures such as C-L and ME include bucket traps, Champ traps, Jackson traps, and Steiner traps. Research has been conducted in South Korea on non-target insects captured in traps used for Tephritid fruit flies, as well as on innovative dispensing systems for male lures intended to detect invasive fruit flies [23-27].

In many regions, male annihilation technique (MAT) carriers, such as cotton wicks, molded paper fiber, fiberboard blocks, and Min-U-Gel are commonly employed. For example, fiberboard blocks infused with ME and organophosphate insecticides like naled and malathion have been used in Okinawa to control *B. dorsalis* and in Australia to eliminate the papaya fruit fly, Bactrocera papaya. Traditionally, liquid lures, often a blend of ME or C-L and insecticides like malathion or naled, are placed on cotton wicks. However, these liquid formulations require careful handling and pose potential health risks due to pesticide exposure [28-32].

In recent years, there has been a shift towards using solid formulations of lures such as C-L plugs, ME cones (Scentry), and ME wafers (Farma Tech) in place of liquid lures and insecticides. These solid lure/insecticide combinations, such as DDVP, have proven to be more convenient and safer for field workers [33, 34]. Studies have shown that traps without insecticide and only a male lure generally capture fewer males of species like *Z. cucurbitae* or *B. dorsalis* compared to those with a lure plus naled or a separate DDVP strip. Additionally, experiments demonstrated that adding spinosad, a low-risk pesticide, to the traps did not significantly increase their effectiveness over traps with just the male lure [35-38].

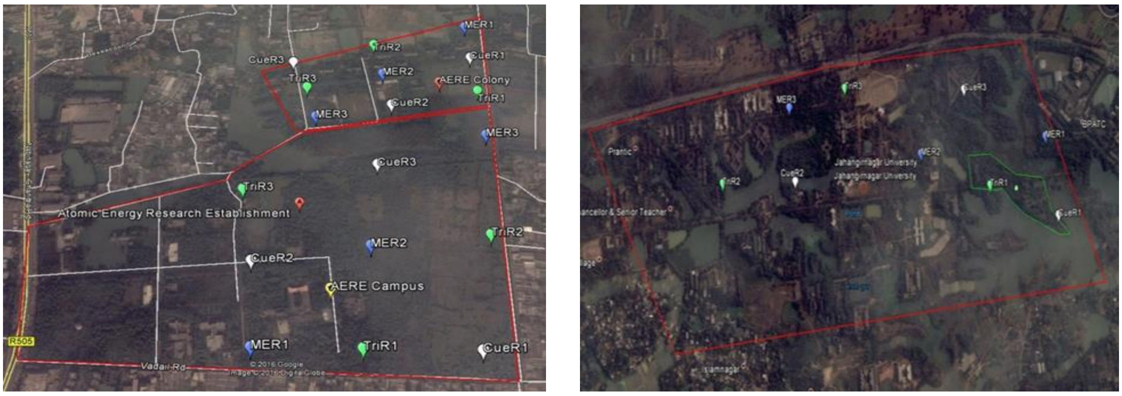
Despite the move towards safer alternatives, organophosphate insecticides are still commonly used in fruit fly surveillance programs to ensure effective trapping. Pre-packaged DDVP strips, which are easier and safer to handle than solutions combining lure and naled, are just as effective for monitoring Tephritid populations and detecting infestations. The Hawaii Fruit Fly Area-Wide Pest Management Program (HAWPM) (2000-2009) stands as a prime example of successful research and development of fruit fly monitoring and control technologies, including monitoring, field sanitation, protein bait sprays, MAT, parasitoid releases, and sterile insect releases.

However, the use of male lures in large-scale fruit fly eradication programs has raised concerns about potential non-target effects on beneficial insects. These concerns suggest that such programs may inadvertently harm non-target species or even threaten small, endemic insect populations, highlighting the need to be careful [39-41].

In Bangladesh, the discovery of a new species and 33 additional records of Tephritid fruit flies has been documented. Four species—*B. dorsalis, Z. tau, B. zonata,* and *Z. cucurbitae*—are particularly harmful to the production of fruits and vegetables. Recently, pheromone traps have become an essential tool for monitoring these pests across various crops in the country. The effectiveness of these traps is influenced by factors such as the trap design, placement, and chemical composition used. Additionally, the development of various lures, the introduction of new lures, and the combination of lures with traps are key factors for capturing pest fruit flies. However, there is limited research on the use of solid male lure formulations and the impact of these traps on non-target and beneficial insects in Bangladesh [42-46]. This study aims to assess the effectiveness of 3 solid single lure plugs (ME, C-L, and TML) combined with insecticide strips (DDVP) in trapping four economically significant dacine fruit fly species in Bangladesh, while also examining their effects on non-target insect attraction.

Materials and Methods

Study locations
The study was conducted in three green zones of Bangladesh from May to September 2015, where the capture of 2 cucurbit pests, *Z. tau*, and *Z. cucurbitae*, along with fruit pests *B. dorsalis* and *B. zonata*, was observed. The experimental sites **(Figures 1a and 1b)** included: i. The Atomic Energy Research Establishment (AERE) colony, Savar, Dhaka (8.64 ha) at coordinates 23°57'35.60" N, 90°16'54.02" E; ii. The AERE office campus, Savar, Dhaka (112.276 ha) at coordinates 23°57'14.62" N, 90°16'44.79" E; and iii. Jahangirnagar University (JU) campus (214.62 ha) at coordinates 23°52'8.85" N, 90°16'1.50" E. The mean monthly rainfall in these areas was 394.5 mm (ranging from 185 mm to 623 mm), with a mean temperature of 29.17 °C (minimum 25.7 °C, maximum 31.8 °C) and average relative humidity of 77% (ranging from 71-81%). These locations featured a mix of agricultural fields, backyard gardens, and plantations with a variety of fruit trees (such as jackfruit, guava, mango, oranges, starfruit, and banana) and vegetables (including melon, pumpkin, brinjal, and chili peppers), alongside other non-host trees. These sites are representative of the typical agricultural environments in Bangladesh, where dacine fruit flies are commonly found.



a) b)

Figure 1. a) Traps locations at AERE colony (23°57'35.60" N, 90°16'54.02" E), AERE office campus (23°57'14.62" N, 90°16'44.79" E), b) JU campus (23°52'8.85" N, 90°16'1.50" E), (average temperature 30 ˚C and precipi. 55-601 mm).

Lure and trap setup
In this study, three types of solid single lure plugs were used: i. C-L, ii. ME, and iii. TML (Scentry Biologicals, Billings, Montana, USDA-APHIS-PPQ), along with DDVP strips (10% dichlorvos, Vapertape® II, Hercon Environmental, Emingsville, Pennsylvania, USA). These were placed in traps across the experimental sites. Traps were positioned at nine different locations within each of the 3 study areas, suspended from tree branches approximately 1.5 meters above the ground in shaded spots using metal hangers. The traps consisted of 1/2 liter plastic containers with two 10 mm round holes near the top for fly entry. Fly collections occurred weekly for a total of 18 weeks. Each week, traps were emptied, and all captured flies and non-target arthropods were collected in plastic bags and transported to the Insect Biotechnology Division (IBD) laboratory at AERE for counting. Flies of the Zeugodacus/Bactrocera species were identified to the species and recorded in an MS Excel spreadsheet. To reduce positional bias, the traps were rotated clockwise each week within their respective areas. Weather data, including rainfall and temperature, were obtained from the Bangladesh Meteorological Department, Dhaka, Agargaon, Bangladesh.

Statistical analysis
The data were analyzed using ANOVA (Analysis of Variance) through Mini-Tab statistical software (version 2017, USA). Differences between treatment means were assessed using the Tukey HSD Test at a 0.05 significance level.

Results and Discussion

During the trapping study in three green spaces of Savar, Dhaka, Bangladesh, a total of five dacine fruit fly species (*Z. cucurbitae*, *Z. tau*, *B. dorsalis*, *B. zonata*, and *B. nigrofemoralis*) along with one *Dacus* species *D. longicornis* were captured. Among these, *B. dorsalis* was the most abundant, accounting for 89.56% of the captured specimens at the Jahangirnagar University (JU) campus, where ME baited traps captured an average of 1614.1±14.9 flies over the 18 weeks (538.05±62.28 flies per trap per week (FTW)). *Z. cucurbitae* (138.0±21.83 FTW) and *Z. tau* (35.11±7.13 FTW) were predominantly trapped by C-L baited traps at the AERE office campus and JU campus, respectively. Interestingly, TML-baited traps failed to attract any *Bactrocera* or *Zeugodacus* species, suggesting no presence of *Ceratitis* or *Anastrepha* species in the trial areas during the experiment.

The number of *Z. cucurbitae* captured varied significantly between campuses (df = 2, 51; F = 6.12; P = 0.004). While no significant difference was found between the AERE office campus (138.0 ± 21.83 FTW) and the JU campus (122.44 ± 13.53 FTW), the AERE colony recorded significantly lower captures (62.25 ± 11.20 FTW) compared to the other two sites. The capture of *Z. tau* (35.11 ± 7.13 FTW) was also significantly higher at the JU campus (df = 2, 51; F = 13.64; P = 0.000), while the AERE office campus (7.83 ± 1.86 FTW) and AERE colony (6.30 ± 1.82 FTW) showed much lower, statistically insignificant captures.

B. dorsalis capture was highest at the JU campus (538.05 ± 62.28 FTW) (df = 2, 51; F = 33.32; P = 0.000), followed by the AERE office campus (204.69 ± 37.07 FTW) and AERE colony (64.00 ± 9.20 FTW), with significant differences between locations. *B. zonata* was captured in minimal numbers across all sites, with no statistical differences (df = 2, 51; F = 2.89; P = 0.065), ranging from 0.77 ± 0.37 to 4.06 ± 1.28 FTW.

A comparison of the total captures of Bactrocera and Zeugodacus species at the AERE campus revealed significant variation (df = 3, 68; F = 21.27; P = 0.000). The highest number of *B. dorsalis* (204.7 ± 157.2 FTW) was captured, followed *by Z. cucurbitae* (138.1 ± 92.6 FTW), *Z. tau* (7.83 ± 7.9 FTW), and *B. zonata* (4.06 ± 5.4 FTW). At the AERE colony, similar results were observed, with *B. dorsalis* (64.00 ± 39.07 FTW) being most abundant, followed by *Z. cucurbitae* (62.3 ± 47.6 FTW), *Z. tau* (6.31 ± 7.7 FTW), and *B. zonata* (4.0 ± 5.7 FTW) (df =3, 68; F = 20.80; P = 0.000). At the JU campus, *B. dorsalis* was by far the most captured species (538.05 ± 62.28 FTW), followed by *Z. cucurbitae* (122.4 ± 57.2 FTW), and *Z. tau* (35.11 ± 30.28 FTW). The capture of *B. zonata* remained the lowest (0.7 ± 1.5 FTW) among all species.

Regarding non-target insects, 132, 1304, and 21 were captured in C-L, ME, and TML-baited traps at the AERE colony, AERE office campus, and JU campus, respectively. The most common non-target species in the traps were Drosophilidae, Hymenoptera (black ants), Milichiidae, and Muscidae, which were particularly attracted to the decaying fruit flies in the traps. Control traps, however, captured very few non-target insects. Scavenger species were primarily observed in the traps across all locations **(Table 1)**.

Table 1. Non-target insect capture in traps baited with ME, C-L, and TML, compared to control traps, across three experimental fields in the Savar area (May to September 2015).

Experimental fields	Order/Family/Genus/Species	Cue-lure (C-L)	Methyl-eugenol (ME)	Trimed-lure (TML)	Control
AERE colony	Coleoptera (beetles)	-	-	-	-
	Drosophilidae	0.3 ± 0.1	0.16 ± 0.1	-	-
	Hymenoptera (black ants)	1.66 ± 0.7	0.27 ± 0.6	0.11 ± 0.1	-
	Lonchaeidae	-	1.16 ± 2.5	-	-
	Milichiidae	0.11 ± 0.1	0.11 ± 0.1	0.96 ± 0	-
	Muscidae (Atherigona)	0.55 ± 1.1	0.33 ± 0.5	0.33 ± 0.5	-
	Platystomatidae (*Agadasys hexablepharis*)	0.01 ± 0.1	-	-	-
AERE office campus	Arachnids (jumping spiders-Salticidae)	0.33 ± 0.2	-	-	-
	Braconidae	-	10.33 ± 14.6	-	-
	Coleoptera (beetles)	14.0 ± 33.0	4.2 ± 11.0	0.55 ± 0.1	-
	Drosophilidae	2.94 ± 1.7	1.6 ± 0.9	0.2 ± 0.1	-
	Hymenoptera (Pompilidae, weaver ants)	-	1.5 ± 2.0	-	-
	Lepidoptera (moths)	10.33 ± 14.6	21.6 ± 47.2	-	-
	Orthoptera (grasshoppers)	1.4 ± 1.8	7.6 ± 7.8	-	-
	Milichiidae	0.01 ± 0.1	0.16 ± 0.5	-	-
	Muscidae (Atherigona)	0.05 ± 0.1	0.01 ± 0.1	-	-

	Platystomatidae (*Agadasys hexablepharis*)	0.1 ± 0.1	0.01 ± 0.2	-	-
	Sarcophagidae	21.6 ± 47.2	0.16 ± 0.5	-	-
JU campus	Arachnids (jumping spiders - Salticidae)	0.16 ± 0.1	0.22 ± 0.0	0.16 ± 0	-
	Bugs	0.72 ± 0.4	0.66 ± 0.2	0.60 ± 0.1	-
	Coleoptera (beetles)	0.55 ± 0.1	3.22 ± 2.1	3.16 ± 0.5	-
	Drosophilidae	3.0 ± 0.7	-	-	3.0 ± 0.7
	Hymenoptera (black ants)	0.16 ± 0.5	3.16 ± 0.5	5.16 ± 1.6	-
	Lepidoptera (moths)	5.16 ± 1.6	-	-	-
	Milichiidae	1.5 ± 2.0	-	-	-
	Muscidae	1.5 ± 2.0	2.94 ± 3.6	-	-
	Platystomatidae (*Agadasys hexablepharis*)	0.83 ± 0.8	-	-	-
Total	Non-target insects captured	132	1304	21	-

In this study, the capture of *B. dorsalis* was significantly higher than *Z. tau*, *Z. cucurbitae,* and *B. zonata* when traps baited with solid lure plugs of ME and C-L were placed at the AERE office campus. These findings share partial similarities with the results of Hossain *et al*. [44], who reported that *B. dorsalis* was the predominant polyphagous fruit fly species (58.0%), followed by *Z. cucurbitae* (23.6%) and *Z. tau* (13.5%), based on traps baited with solid lure plugs of ME, C-L, and zingerone during a two-year survey at the same campus. Our experiment, which utilized ME, C-L, and TML baited traps over 18 weeks, found that the overall capture rate of dacine fruit flies was greater at the JU campus compared to the AERE office campus and AERE colony **(Figures 2-4)**.

In contrast, the common parahormone lure stick, widely used by fruit and vegetable growers in Bangladesh, consists of a small cotton wick or rope impregnated with 2 ml of lure. A cotton ball soaked with a 4% Sevin solution (a contact poison from ACI Limited, Bangladesh) is placed inside the trap to both lure and kill the flies. A mango orchard experiment in Chapai Nawabganjj showed that traps baited with solid single ME lure plugs (from Scentry Biologicals, Billings, Montana, USDA APHIS-PPQ) captured more *B. dorsalis* and *B. zonata* than traps using commercially available ME-impregnated cotton rope/wick plugs from Ispahani Co. Ltd. (Bangladesh) (unpublished data).

Additionally, solid ME lure plugs have been employed to study the population trends of male *B. dorsalis* and to assess the abundance of *B. zonata* in mango orchards. Previous field studies have shown that traps baited with either solid or liquid male lures yield similar capture rates of *Bactrocera* males. Despite this, several studies advocate for alternative lure delivery systems. However, it remains common practice in such trapping methods to include insecticides (such as nailed or DDVP or other contact poisons) alongside the male lure.

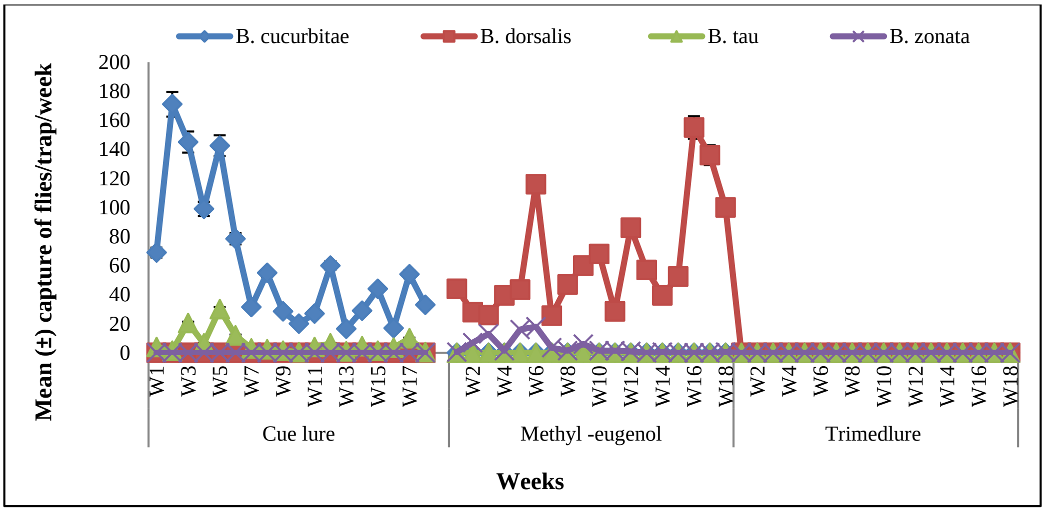


Figure 2. Average (± SE) weekly capture rates of *B. zonata*, *B. dorsalis*, *Z. tau*, and *Z. cucurbitae* using three types of solid lure plugs (C-L, ME, and TML) along with insecticide strips in traps at AERE colony from May to September 2015.

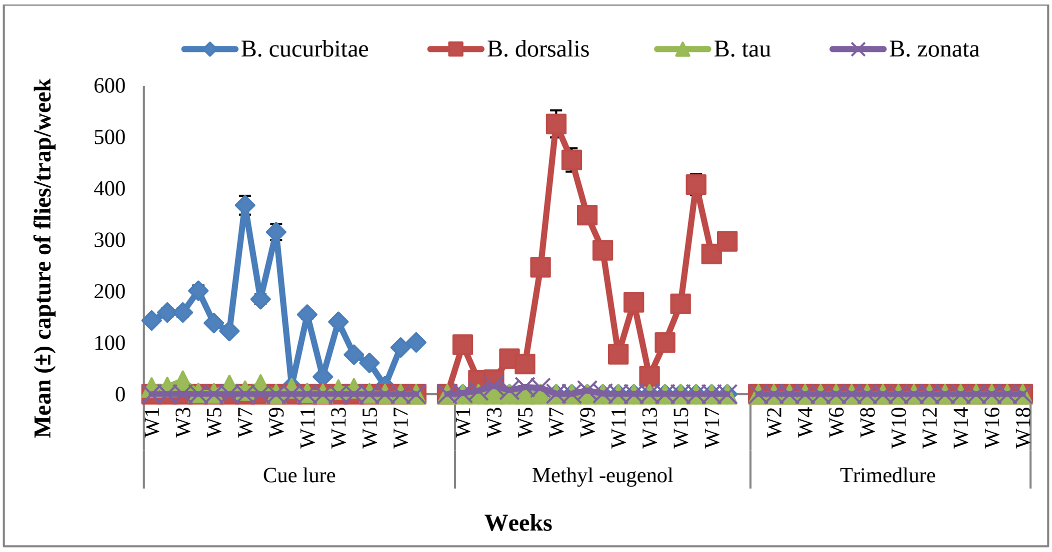


Figure 3. Average (± SE) weekly capture rates of *B. zonata*, *B. dorsalis*, *Z. tau*, and *Z. cucurbitae* per week in traps baited with three solid lure plugs (C-L, ME, and TML) and insecticide strips at AERE office campus during May-September 2015.

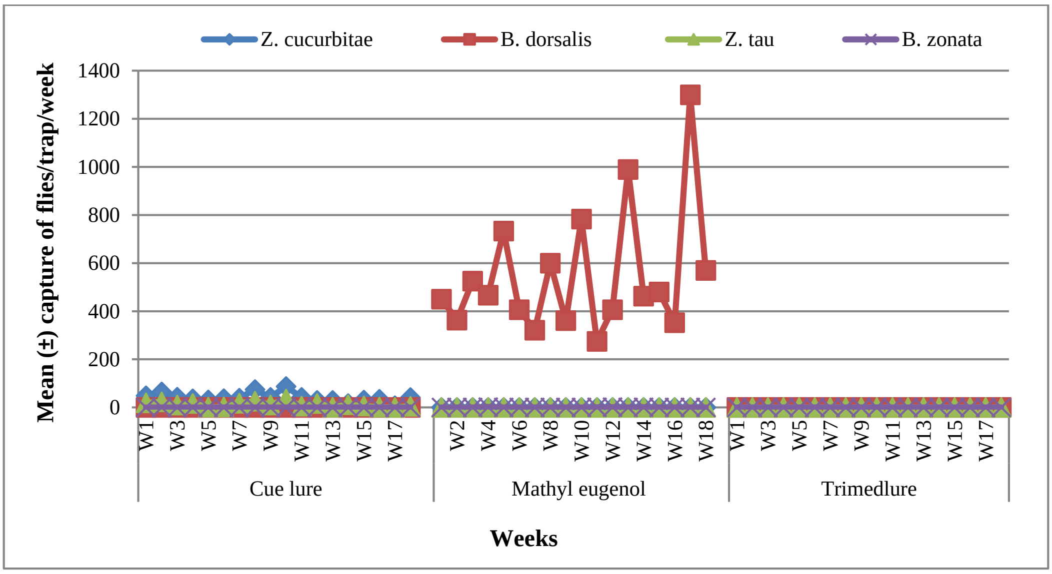


Figure 4. Mean (± SE) weekly capture rates of *B. zonata*, *B. dorsalis*, *Z. tau*, and *Z. cucurbitae* per week in traps baited with three solid lure plugs (C-L, ME, and TML) and insecticide strips baited traps at JU campus from May to September 2015.

The findings of this study regarding the capture of non-target insects in traps with different lures show some similarities with the work of Leblanc *et al*. [47, 48], who documented a wide variety of non-target species, with Drosophilidae, Sarcophagidae, Chloropidaem, Ceratopogonidae, Calliphoridae, Neriidae, Cecidomyiidae, Muscidae and Corylophidae being the most frequent in traps baited with multiple lures for Tephritid fruit flies in Hawaii. Similar trends were seen in the current study, which also confirmed the attraction of scavenger species to food lures and decaying fruit flies in traps using male lures [26, 47-49]. However, the number of non-target insects captured here was notably lower than in previous studies, likely because synthetic lures were used instead of food-based ones. The relatively short duration of the study (18 weeks, from May to September 2015) is another factor that limits the applicability of these results to specific weather conditions and host availability during that year. It's possible that results could differ in other seasons, such as autumn or the cooler months of winter.

Conclusion

This study highlights the species-specific effectiveness of solid lure plugs in attracting dacine fruit flies. The combination of solid lures and DDVP insecticide strips was not only efficient for mass-trapping but also user-friendly, with minimal disruption to non-target beneficial insects. While some non-target species were drawn to the traps, their presence was largely due to decaying Bactrocera flies rather than the lures themselves. The overall impact of male lures on non-target insects appeared to be insignificant. Moving forward, research should prioritize the development of alternative lure matrices derived from natural sources, refining lure formulations, and identifying safer insecticides to improve fruit fly management. Such advancements will support more sustainable control strategies, integrating lure-based detection with protein bait sprays, proper sanitation, and eco-friendly methods like the Sterile Insect Technique within the Area-Wide Integrated Fruit Fly Management Program (AW-IFFMP) in Bangladesh.

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Conflict of Interest: None

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Ethics Statement: None

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