Chapter 16
Use of Para-pheromone Methyl Eugenol for Suppression of the Mango Fruit Fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae) in Southern Ethiopia

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Abstract Mango growers in Southern Ethiopia are faced with the severe challenge of controlling mango fruit fly, *Bactrocera dorsalis*. Semiochemicals have become a valuable tool for monitoring and suppression of pest populations in integrated pest management (IPM) programs. Since pheromone traps are relatively easy to use, cost-effective, species-specific and environmentally friendly tools for IPM programs, we evaluated the performance of methyl eugenol (ME) vs. *B. dorsalis* in Gamo-Gofa zone, in Southern Ethiopia. A 3-month (February–April 2018) trial on *B. dorsalis* population reduction was implemented in six intervention sites of Arba minch zuria district, and three control sites of Mierab Abaya district. Results showed that continuous application of ME was effective in reducing the fruit fly population. Fruit fly captures and fruit infestation in six intervention sites were significantly lower during the 3 months than those recorded in the three control sites. Therefore, the strategy of including mango fruit fly suppression techniques using pheromone and/or parapheromone lures such as ME (related to mating behaviour) in IPM approach, is recommended.

Keywords Mango · Mating disruption · Fruit fly · Horticulture

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16.1 Introduction

Mango is one of the most important fruit crop produced and exported in Ethiopia. In the recent years, its production showed a steady increase in the planted acreage. Like other perennial fruit crops, mango production is plagued with many insect pests. In Africa, of the 1.9 million tons of mangoes produced annually, about 40% is lost due to fruit flies, where infestation rates vary among countries and seasons, ranging from 5 to 100% (Lux 1999; Goergen 2011).

Across Southern and Eastern parts of Ethiopia, *Ceratitis* spp. (*C. capitata* and *C. fasciventris*) and *Bactrocera* spp. (mostly *B. dorsalis*), are the dominant species, with a great economic importance across the country (Ferdu et al. 2007; Dawit et al. 2015). However, the most important fruit flies on which efforts of pest management are concentrated is the mango or Oriental fruit fly, *B. dorsalis* Hendel (Diptera: Tephritidae).

In the recent past, Ekesi et al. (2009, 2010) reported that *B. dorsalis* competes with other endemic fruit fly species such as *C. cosyra* Walker, *C. capitata* Wiedemann and *C. ditissima* (Munro) causing species displacement. It is estimated that where plantations are not managed, there could be total fruit loss, although losses less than 30% can be salvaged if control measures are applied. *Bactrocera dorsalis* was first detected in Kenya in 2003 (Lux et al. 2003; Drew et al. 2005; Ekesi and Billah 2009) and in Ghana in 2005 (Billah et al. 2006). It is a serious pest, requiring the adoption of area-wide control measures to suppress the populations below their economic threshold.

Several control measures against fruit flies have been practised including, among others, chemical control. Some of the most widely used control measures which pose little or no harm to the environment is the use of traps and semiochemicals such as sex pheromones, lures and baits (Cunningham et al. 1978; Agunloye 1987; McQuate et al. 2005). These are generally used for masse trapping, spot spray and in “attract and kill” approaches. The success of the mass trapping strategy depends on the efficiency of traps and lures (Cohen and Yuval 2000). Trap designs, including assorted colours and shapes, can also influence efficacy in fruit fly catches (Epsky et al. 1995; Vargas et al. 1997).

Traps baited with sex pheromones attract males of the same species for mating. They have become a valuable tool for monitoring and suppression of fruit fly pest populations in survey and integrated pest management (IPM) programs. Many insect sex pheromones can now be chemically synthesized for use in pest monitoring and mating disruption. Since pheromone traps are relatively easy to use, cheap, species-specific, and environmentally benign, they make ideal tools for IPM programs.

For tephritid fruit fly suppression, methyl eugenol (ME) and cue-lure are highly attractive kairomone lures to *B. dorsalis* and the melon fly, *B. cucurbitae* (Coquillett), respectively.

The effectiveness and performance in the field of ME-based baits against mango fruit fly in Ethiopia have not yet been documented in the literature, though other studies have suggested its effectiveness, elsewhere.

The International Centre of Insect Physiology and Ecology (ICIPE-Ethiopia) initiated an area-wide fruit fly suppression program through pheromone traps in Gamo-Gofa zone of Southern Regional State of Ethiopia, since 2017. Therefore, the purpose of this study was to demonstrate and validate the performance of solid male lure ME formulated with killer insecticide for suppression of mango fruit flyin mango fields, as alternatives to current control systems based on organophosphate insecticides.

16.2 Materials and Methods

16.2.1 Description of Study Sites for Intervention and Control

The study was conducted in Arba Minch Zuria and Mierab Abaya Districts of Gamo-Gofa zone, in the Southern Regional State of Ethiopia. The two districts are located at a grid reference between (5° 50.46′ N and 37° 27.72′ E) and (6° 27.32′ N and 37° 44.64′ E), respectively. Six villages (Ankober, Wajifo, Kolashele, Elgo, Chelba and Lante) in Arba Minch Zuria District were selected for intervention, while three villages (Ugayehu, Molie and Kolamulato) in Mierab, Abaya District, were selected for control purposes. The GPS coordinates, temperature and rainfall of all nine locations are shown in Table 16.1. These two Districts and nine villages are known for major mango production in the zone. Majority of the inhabitants of these areas are involved in subsistence agriculture cultivating mango, banana, apple, avocado, papaya and guava.

Table 16.1 GPS position, altitude, temperature and rain fall of study sites

Location	GPS Position	Altitude (mamsl)	Temperature (°C)	Rainfall (mm)
Ankober	6° 14.581′ N, 37° 44.680′ E	1209	19–32	595.5
Wajifo	6° 27.320′ N, 37°44.643′ E	1220	18–34	610
Kolashele	5° 52.919′ N, 37° 29.740′ E	1162	17.6–31.6	588.4
Elgo	5° 50.469′ N, 37° 27.721′ E	1122	20.6–35.6	588.4
Chelba	6° 6.496′ N, 37° 35.074′ E	1203	17.6–31.6	590.4
Lante	6° 7.834′ N, 37° 38.165′ E	1190	17.6–32.6	588.4
Ugayehu	6° 15.417′ N, 37° 45.663′ E	1218	19–34	610
Molie	6° 16.25″ N, 37° 46.367′ E	1215	19–34	603
Kolamulato	6° 27.942′ N, 37° 45.081′ E	1202	18–34	600

Fig. 16.1 Lynfield bucket traps

16.2.2 Treatments for Insect Population Suppression

Fruit fly baits are generally short distance attractants. For this reason, we chose whole orchards for para-pheromone ME attract and kill applications. The attract and kill concept consists in eliminating males from the vicinity, for mating disruption.

The following sites: Ankober, Wajifo, Kolashele, Elgo, Chelba and Lante were treated with the commonly used polymeric plug of solid para-pheromone ME formulated with insecticide malathion, to attract and kill males *B. dorsalis* at monthly intervals, throughout the fruit development period. The treatment interval selected was in line with the manufacturer's recommendation (4–6 weeks), based on research with codling moth (Charmillot et al. 2000). Control sites within a 20–30 km radius (Ugayehu, Molie and Kolamulato) were used as untreated control orchards.

Fruit fly populations were monitored using yellow Lynfield traps, a bucket type trap composed of a cylindrical plastic container with four equidistant holes on the upper third (Fig. 16.1). The lid of the trap contains a hook to which a ME dispenser could be fitted. Intervention traps were maintained on the field for 3 months (February–April 2018). Between 50 and 100 traps were placed in each intervention site, depending on the farm size, at the distance of 50 m apart, hanged at a height of 1–2 m from the ground.

The control villages were at farmers practice. 8 ME baited monitoring traps were deployed in each control site at the distance of 80 m apart. They stayed for 24 h per month, and caught flies were collected at a 1-week interval during the 3 months, and counted.

16.2.3 Insect Population Monitoring

All traps were placed at the rate of ten traps per hectare and placed at un-shaded area to sunlight on the windward side of a field, so that the pheromone should be blown into the field. Traps were coded and numbered. They were managed by individual farmers who were responsible for the traps safety and service. The traps were serviced at weekly interval and caught flies were collected weekly at each service time and visit, counted and data recorded for further analyses.

The caught flies per trap per day (FTD) were also calculated to facilitate comparison across the different intervention and controlled localities.

The formula used to calculate FTD was:

$$\text{FTD} = F / T \times D$$

whereby F = total number of flies; T = number of serviced traps; D = average number of days the traps were exposed in the field.

16.2.4 Assessment of Fruit Infestation

Throughout the study period the fruit fly infestation was assessed by monthly fruit collections from mango orchards. Every month, 5 to 10 kg fruits were sampled from both intervention and control sites. Fruits were weighed and then assessed visually for rotting and presence of maggots. Afterwards, the fruits were sorted into “Infested” and “Non-Infested” categories, counted and weighed again separately. The clean-looking mango fruits were kept in containers for a week then dissected for the presence of fruit fly maggots. The proportions of damaged fruits were added in to “Infested” category to recalculate the level of infestations. Percent infested fruits was determined as ratio of number of infested fruits per total number of collected fruits. The experiment was repeated three times, one month apart.

16.2.5 Fruit Fly Impact Assessment and Statistical Analyses

The impact of the fruit fly on mango at zone level was assessed at the yield and economic levels. Key informant interviews (KIIs) were used to assess the outcome and impact of fruit fly on mango production

16.2.6 Data Analysis

For normalization, data on fruit fly captures and proportion of fruit infestation were log- transformed to (Log10) and angular (arcsine $\sqrt{\text{proportion}}$), respectively, for statistical comparisons. Untransformed means are presented in both figures and tables. Analysis of variance was used to determine differences among study sites using SAS Statistical Program. Once a significant difference was detected, data were subjected to post hoc analysis for means separation using LSD test, ($P = 0.05$).

Comparison of fruit fly population levels between intervention and control sites (fruit fly suppression test) was made using t-tests (Sokal and Rohlf 1981) of log-transformed trap catch results by Statistical Package for Social Science (SPSS) software. Comparison of percentage fruit infestation by fruit fly between intervention and control sites was made using paired t-tests of angular transformed proportion damaged data.

16.3 Results

16.3.1 Fruit Fly Populations

The attract-and-kill technique significantly reduced the population of *B. dorsalis*, in mango orchards. Average trap catch data in intervention and control sites of each orchard and trap servicing month are presented in Fig. 16.2. Average trap catch in the intervention sites was lower than in the control section in all months, with catch significantly lower in March and April (Fig. 16.2). Trap catch per day in the control sites were more than 20-fold higher, during the whole period of study (Fig. 16.2).

A continuous decrease in fruit fly population was observed over the 3 months under intervention trial sites (Fig. 16.3). At three of the intervention sites (Lante, Elgo, and Chelba) the *B. dorsalis* populations were the lowest throughout the study time. In general, from the time of the first month until the end of the trial, oriental fruit fly trap catch was not higher than 20 flies/trap/day in any intervention sites except in Ankober which was higher than 45 flies/trap/day (Fig. 16.3). In the first month, trap catch in all intervention sites, except Wajifo and Chelba, was higher

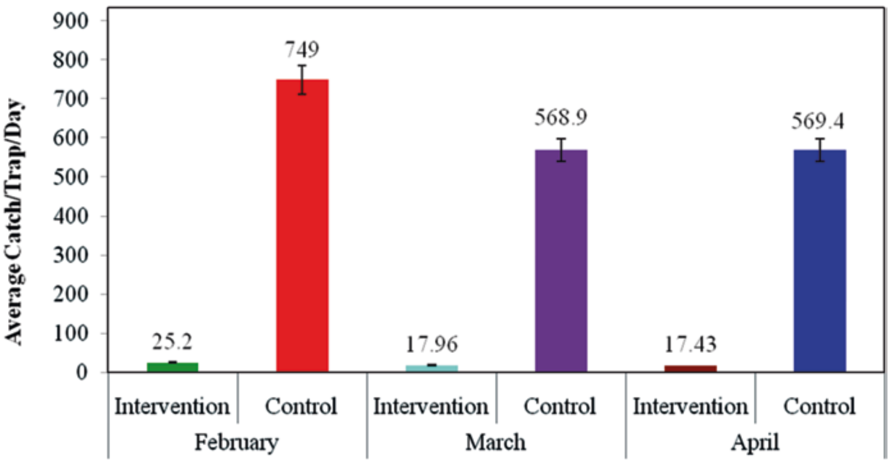

Fig. 16.2 Average catch (flies/trap/day) of *Bactrocera dorsalis* in baited traps with para-pheromone methyl eugenol blend, in intervention versus control sites, at each trap service month

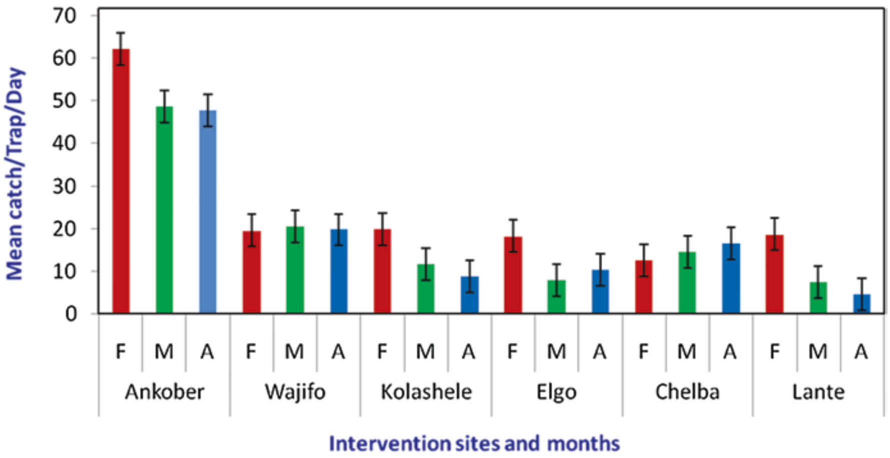

Fig. 16.3 Mean total mango *B. dorsalis* trap catches (flies/trap/day ± SEM) in intervention sites of six villages, at each trap service month

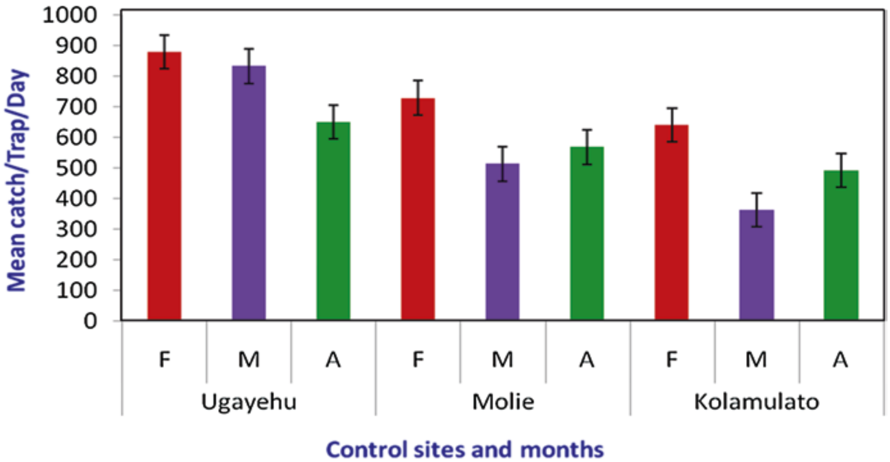

Fig. 16.4 Total mango *B. dorsalis* trap catches (flies/trap/day, ± SEM), in control sites of three villages, at each trap service month

than in the second and third months, although trap catches at second and third months in all intervention sites were not statistically different. The oriental fruit fly populations in the control sites (Ugayehu, Molie and Kolamulato), collected within 24 h of the 3 months were considerably higher throughout trapping periods (Fig. 16.4). Highest fly catches per trap per day were recorded at Ugayehu in February and March (879 and 832 flies, respectively). The lowest catches (362 and 492 flies/trap/day) were observed at Kolamulato in March and April, respectively (Fig. 16.4).

Table 16.2 Mean catches of *Bactrocera dorsalis* males (flies/trap/day ± SEM), from February to April 2018 at Arbaminch and Mierab Abaya districts, Gamo-Gofa zone

Locations	Fruit fly moth catch/trap*
Ankober	52.84 ± 4.69 c
Wajifo	19.95 ± 3.18 c
Kolashele	13.44 ± 3.35 c
Elgo	12.12 ± 3.15 c
Chelba	14.57 ± 1.12 c
Lante	8.25 ± 5.26 c
Ugayehu (control)	786.67 ± 70.16 a
Molie (control)	602.63 ± 64.63 b
Kolamulato (control)	498.00 ± 80.31 b
Mean	223.16
LSD (5%)	113.18
CV (%)	29.29

*Means followed by the same latter in a column are not significantly different ($P \leq 0.05$)

Average trap catches in both intervention and control locations of each site are shown in Table 16.2. For all intervention and control orchards considered together, there was significant difference in mango fruit fly trap catch between treated and control locations. However, there was no significant difference in trap catch in intervention sites while a significant difference was observed among control sites. At three of the control sites (Ugayehu, Molie, and Kolamulato) mango fruit fly trap catches were significantly higher. The mango fruit fly populations in the Ugayehu site (786.67 ± 70.16), however, were considerably highest and statistically significant than in the other remaining sites (Table 16.2). However, significantly lower flies were caught in the intervention sites. Numerically the lowest trap catches were recorded from Lante (8.25 ± 5.26), but the difference was not statistically significant from the other intervention sites ($P < 0.05$, Table 16.2).

16.3.2 Fruit Fly Infestations

Fruit infestation rates decreased over time in both sites. However, a significant difference was observed in the intervention compared to the control sites (Fig. 16.5). Average fruit fly infestation in the mango fruits collected from intervention sites at each of the treatment interval ranged from 11% to 13%, while the infestation in control sites ranged between 24 and 68.4% (Fig. 16.5). Fruit fly infestation obtained from control site was fivefold higher than infestation obtained from intervention site (Fig. 16.5). Overall there were no significant differences in infestation rates among intervention sites, at any of the collection times, while a significant difference in infestations was observed among fruit collection times. The highest infestation rates in any site with the intervention (13%) and control (68.4%), were found in the first

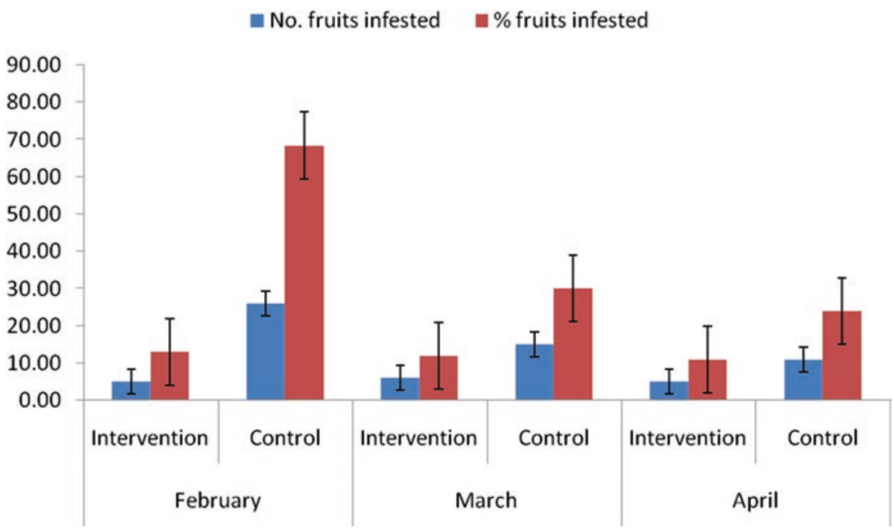

Fig. 16.5 Average (± SEM) number and percentages of fruit fly infestation in intervention versus control sites, at each trap service month

Table 16.3 The extent of mango fruit infestation by fruit flies in different locations of Gamo-Gofa zone, Southern Ethiopia

Location	Number of fruit sampled	Fruits infested (%)∗
Lanate	119	24.23 ± 7.54 c
Chano mille	125	29.85 ± 2.17 c
Kolla shelle	110	27.09 ± 7.97 c
Ugayehu	119	60.91 ± 7.15 a
Molie	125	50.15 ± 8.03 b
Kolamulato	110	43.24 ± 9.38 b
Mean		39.24
LSD (5%)		9.63
CV (%)		13.49

∗Means followed by the same latter in a column are not significantly different ($P \leq 0.05$)

fruit collection month. This month was also had the highest fruit fly population numbers, based on trap catches.

In general, mango infestation levels varied among separate locations of Arbaminch and Mierab Abaya Districts of Gamo-Gofa zone. The average infestation ranged from 24% to 60% in the study locations (Table 16.3 and Fig. 16.6). Significantly higher mango fruit infestations were recorded in Ugayehu, Molie and Kolamulato with respective mean infestations of 60.91%, 50.15% and 43.24% at $P < 0.05$ (Table 16.3). The lowest level of infestation (24.23%) was observed in Lanate (Table 16.3).

Fig. 16.6 Comparison between mango quality at intervention and control sites

Table 16.4 The extent of mango losses by fruit fly in Gamo-Gofa zone during 2017

Parameters	Extent of loss
Yield loss (Qt)	199,620
Yield loss (% of total)	30
Monetary loss (Birr)	159,696,000

NB: Total mango production area in the zone = 2218 ha; mango productivity for 2017 = 300 qt/ha; total mango produced in the zone during 2017 = 665 400 qt; farmgate price of mango in 2017 = 8.00 ETB/kg

16.3.3 Yield and Economic Loss of Mango by Fruit Fly

Table 16.4 summarizes the extent of mango fruit losses due to fruit fly in Gamo-Gofa zone, both in quantity and economic terms. Based on secondary data obtained from the zone and the District Bureau of Agriculture during the year of 2017, damage due to the fruit fly amounted to a yield loss of 199 620 qt (equivalent to 30% of the total mango produced, 665 400 qt) and around 159.7 million Birr (Table 16.4).

16.4 Discussion

Mango fruit flies cause considerable economic damage in the Gamo-Gofa zone of the Southern Region of Ethiopia. We applied mass trapping technique which was one of the most effective strategies for fruit fly management (Aluja 1999; McQuate et al. 2005). Mass trapping consists of the use of traps and baits that release specific volatile substances that attract insects to the trap, in which fruit flies are captured and killed (El-Sayed et al. 2009). However, for some fruit fly species, the use of mass trapping as a control tool depends on the availability of an effective and cheap attractant (Villalobos et al. 2017). If the attractant is not specific, it might lead to failure (Suckling et al. 2016).

Male fruit flies are usually attracted by parapheromones (IAEA 2003). In contrast, lures for attracting female fruit flies into traps are based primarily on food or host lures (Dominiak and Nicol 2010). Our study showed that the use of ME has the potential to minimize population of *B. dorsalis*, and the related impact (Navarro-Llopis et al. 2008). Inclusion in monitoring networks of food-based lures that capture both females and males is useful. ME is a powerful attractant to male *B. dorsalis* (Kafu et al. 2012). In combination with malathion it attracted and killed the male mango fruit flies as such rates that mating was disrupted, effectively reducing the population density. Previous work on the evaluation of ME dispensers on males of *Dacus zonatus* under field conditions showed that it effectively attracted high numbers of males, with a potential for monitoring and control of this pest (Qureshi et al. 1992). Similarly, research with treatments containing ME and malathion (EC50) evaluated nutritional attractants including protein hydrolysate, palm extract, sugar, water and dishwashing liquids and ripe mango, under field conditions. The results showed that mango fruit fly populations were attracted more to the protein hydrolysate than to other treatments (Agarwal and Kumar 1999; Khosravi et al. 2018). It seems that the simultaneous use of hydrolyzed protein in bucket traps along with ME and malathion destroyed significant part of the pest population, disrupting mating and significantly reducing damages. These results confirm our findings.

In general, our results confirmed that the capturing rate of fruit flies showed similar dynamics. In both intervention and control sites and within three consecutive months catched fruit flies were lower in the second and third months than in the first one, even if the population in the control sites were considerably higher, compared to the intervention sites. This decline could be due to regular actions of treating in the first month. Based on results, the highest rate of adult insect capture was related to the treatment of ME and malathion, used during previous years of test implementation. However, in some locations since mango had been cultivated at the desired location mixed with avocado, banana, coffee and citrus tress (that are one of the favourite hosts of the mango fruit flies), the remaining population could have survived on these hosts re-starting the populations during the mango seasons. For this reason, it is advisable to avoid mixed planting of mango with other tropical fruits, to prevent damage by polyphagous fruit flies.

The ME in the traps worked quite well in attracting male flies, when serviced weekly to increase the pest control ngconfirming previous findings (Samuel et al. 2016). Our results also agree with Asquith and Kido (1994) and Howarth and Howarth (2000) as placing the lure and toxicant at 1–2 m above the ground in the uncovered canopy was more effective in controlling the fly populations. Our data further suggest that attractants placed closer than 50 m apart will interfere with each other and would be less cost-efficient.

Results showed a significant reduction in fruit infestation in the intervention compared to the control sitesas ME led to a reduction in losses by attracting male flies and impairing mating.

Infestation levels vary among seasons, countries, regions, agro-ecological areas and cultivars (Vayssières et al. 2009), however fruit flies are still a severe limiting factor for crops on a continental scale. Lux et al. (2003) reported that, of the 1.9 million tons of mangoes annually produced in Africa, about 40% was lost due to fruit flies.

In addition to effectively controlling *B. dorsalis* and fruit damage in mango orchards, the attract-and-kill bait stations caused less harm to non-target insects than conventional insecticides. In conventional control, which depends mainly on the application of broad-spectrum insecticides, non-target insects are exposed to acute and long-term toxic effects of insecticides, both directly via contact and/or indirectly, via ingestion of contaminated preys (Bostanian et al. 2009). In attract-and kill systems, the use of insecticides (the killing agent) is limited to the treated devices. Consequently, attract-and-kill bait stations have minimal lethal and sublethal effects on non-target insects and other invertebrates. The use of a pheromone should benefit further by increasing the bait station selectivity, maximizing the number of flies attracted and the dispensers lifespan (Hafsi et al. 2015).

16.5 Conclusions

Studies on fruit flies continue to increase and provide useful knowledge for researchers working in the areas of monitoring and control tactics. So far, there has been an emphasis on chemical control research, especially the use of organophosphates. However, the continued use of insecticides is increasingly limited, making it necessary to evaluate other control strategies for inclusion in fruit fly management. The use of a mass-trapping method using ME reduced mango fruit flies effectively. Therefore, it can then be a candidate technique to replace aerial treatments with synthetic insecticides, applied to suppress this pest. Even if the number of potential trapping targets by this para-pheromone lure is smaller, as only males are strongly attracted, its impact on consequent progeny reduction by mating disruption is very high. Our study demonstrates that an attract-and-kill method using solid ME lure formulated with an insecticide was effective for suppression of the *B. dorsalis* populations in mango orchards. These results indicate that attract-and-kill methods using ME represent a suitable alternative to conventional insecticide sprays for the control of *B. dorsalis*. Therefore, this product can be used in conjunction with other environment-friendly, area-wide IPM programmes, such as sanitation and protein bait sprays, for management of mango fruit fly. This study provides fact-based information and evidence to end-users and policymakers, to facilitate the application of this bait at local markets to control mango fruit flies in Ethiopia.

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