Addition of mango essence to methyl eugenol more attracted to *Bactrocera dorsalis* Complex (Diptera: Tephritidae) on mango plantation in Majalengka, West Java

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Abstract
Addition of mango essence on methyl eugenol was evaluated for its effect on the performance of methyl eugenol in attracting B. dorsalis complex species using plastic bottle traps on mango plantation in Majalengka, West Java from September to December 2016. Various concentrations of mango essence were added to 0.2 mL methyl eugenol as treatments. The results showed that the addition of mango essence on methyl eugenol increased the number of fruit flies trapped. The effective combination was 0.2 mL methyl eugenol + 0.8 mL mango essence. This combination attracted more wild fruit flies than other combinations.

Based on identification, species of fruit flies trapped were B. dorsalis, B. carambolae-B. umbrosa. This study indicated that the aroma of mango essence increases the use of methyl eugenol in monitoring or reducing B. dorsalis complex population on mango plantation.

Keywords: Attractant, *Bactrocera dorsalis* complex, mango essence, methyl eugenol.

Introduction

Mango (*Mangifera indica* L.) is one of the potentially valuable Indonesian fruits for local and international market. According to Badan Pusat Statistik[1], the average production of mango in season 2013 was 2.058.609 tons, higher than that of guava (170.810 tons), papaya (871.282 tons), star fruit (71.431 tons), mangosteen (118.909 tons), and citrus (1.411.229 tons). Several centers of mango plantations in Indonesia, in West Java are Indramayu, Cirebon, Majalengka and Sumedang.[2] There are five common mango varieties such as harum manis, golek, manalagi, cengkir, and gedong gincu[3]. Anisah[4] reported that market demand for mango throughout year 2000 had been continuing to increase which was estimated to reach 533.000 tons for local market, and 114,552 tons for export market to Taiwan, Singapore, and Hongkong.

Sarwono[5] found that one of the obstacles faced by mango fruit farmers in Indonesia to provide the export needs was the pests attack. It caused the quality of mangoes did not meet the standards set by the importer countries. For example, Indonesia could not export mangoes to Japan because fruit fly eggs and larvae were found in the mango fruits. Tepritid fruit flies (*Bactrocera* spp.) are one of the serious pests of mango[6]. The attack can lead to production losses of 30% - 40%, may even reach 100% if the environment condition supports the development of the fruit flies[7]. There are 66 species of fruit flies that have been identified and spread throughout Indonesia. Among these species, several particularly most destructive species of fruit production and quality are the *Bactrocera dorsalis* Complex[8].

Various efforts to control fruit flies have been developed including the use of synthetic insecticides. However, the unwise use of synthetic pesticides causes pollution that is very harmful to the environment and health, so there is a need to control and limit the use of synthetic pesticides and reduce pollution caused by pesticide residues[9]. Therefore, it is necessary to control the fruit fly with environmentally friendly technique by using para-pheromone traps[10].

Among para-pheromones that attract male fruit flies are methyl eugenol, cue lure, trimed[11] and 4- (4-hydroxy-phenyl) -2- Butanone[12]. Each para-pheromone attracts different fruit fly species. Methyl eugenol is a common para-pheromone used in Indonesia, because it attracts *B. dorsalis* and *B. carambolae* which are the highest abundant species in Indonesia[13,14]. Methyl eugenol is needed by male fruit flies to produce sex pheromones. Application of methyl eugenol trap decreases the number of fruit flies as it minimizes the possibility of copulation[15].

The weakness of using methyl eugenol is it only attracts male fruit flies[16]. So, it is necessary to add compounds or substances that can attract female fruit flies. Rattanapun et al[17] found that female fruit flies select their host by using the sense of smell, sight and contact cues such as the color, size, and aroma of the fruit of the host plant. The addition of mango essence to methyl eugenol is expected not only to increase the number of male fruit flies trapped, but also to capture the female fruit flies. Therefore, research of addition of mango essence to methyl eugenol is necessary to improve the performance of fruit fly traps.

Material and Methods

Preparation of methyl eugenol and fruit essence. The methyl eugenol used was obtained from clove distillation, and then it was methylated to methyl eugenol. The distillation and methylation processes were carried out at

Plant Pests Laboratory, Department of Plant Pests and Diseases, Faculty of Agriculture, Universitas Padjadjaran. The synthetic mango essence was obtained from local market.

Preparation of traps. The trap was made by modifying plastic bottle. The upper side of the bottle (± 7 cm from the top) was perforated, four holes, as the entry of the fruit flies. The bottle cap was punctured to hold wire used to hook cotton wick, and also to hook the trap on the mango tree. Methyl eugenol was applied on one side of the cotton wick and mango essence on the other side. The treated cotton was wrapped in a zipped plastic bag with holes for spreading the aroma of methyl eugenol and fruit essence. The trap was added with 200 mL of water and 1% formalin.

Trials and observation. The experiment was conducted at mango plantation in Bantrangsana village, Panyingkiran district, Majalengka regency from September to December 2016. The mango plantation area was 1 ha with approximately 100 mango trees. The experiment was arranged in the randomized complete block design consisting of 6 treatments and 4 replications. The treatments were: (1) 0.2 mL methyl eugenol; (2) 0.2 mL methyl eugenol + 0.2 mL mango essence; (3) 0.2 mLmethyl eugenol + 0.4 mL mango essence; (4) 0.2 mL methyl eugenol + 0.6 mL mango essence; (5) 0.2 mL methyl eugenol + 0.8 mL mango essence and (6) 0.2 mL mango essence.

The traps were installed per line in accordance with the number of replicates and distance between traps was 20 meters. Number of traps installed were 24 units. The traps were installed by attaching wires to branches of trees at 1.5 - 2 meters above ground level. Observation was carried out every month. The number of male and female fruit flies trapped was counted and these wild fruit flies were identified. Biotic data, such as host availability and abiotic data, such as temperature, rainfall, and number of rainy day were also observed to determine factors that may affect the population density of the fruit fly.

Data Analysis: The data were tested for normality using the Kolmogorov-Smirnov test. Abnormal data were transformed, and then analyzed by using analysis of variance (ANOVA) to compare the results between treatments. If the difference in treatments had an effect, then the test was continued with Duncan Multiple Range Test at 5% level. The relationship between the number of fruit fly flies and the abiotic factors was analyzed by using Pearson's parametric correlation and regression analysis. All data analysis was carried out by using Statistical Product and Service Solutions (SPSS) program version 22.0.

Results and Discussion

Effect of addition of mango essence to methyl eugenol in attracting fruit fly: The number of fruit flies trapped were varied each month (table 1). The results showed that the number of male fruit flies trapped was higher than that of female fruit flies. The highest number was in October, and the lowest number was in November.

The results showed that addition of mango essence to methyl eugenol traps gave a positive effect, which could attract more fruit flies than control (table 2). The treatment of 0.2 mL methyl eugenol was able to attract 3640 fruit flies in total during the experiment, whereas treatments with mango-essence addition were able to attract up to double of the control result. The results from September to November showed that treatment of 0.2 mL methyl eugenol + 0.8 mL mango essence was the best treatment in attracting fruit flies whereas in December, the treatment of 0.2 mL methyl eugenol + 0.6 mL of mango essence was the best treatment in attracting fruit flies. This current result indicated the highest concentration of mango essence did not always show the best results every month.

Methyl eugenol is classified as food lure because male fruit fly comes for food purposes[18]. Furthermore, the process of metabolism takes place within the body of the male fruit fly to produce a pulling agent (sex pheromone). Tan and Nishida[19] reported that male fruit flies consuming methyl eugenol were proved to have a stronger competitiveness in mating competition than those which did not. Therefore, methyl eugenol is used to attract the male fruit fly in a trap. On the other hand, Szentesi et al[20] reported that extracts of main host plants may be used to attract fruit fly as chemical stimuli in recognizing the site of oviposition.

Table 1
Number of fruit flies trapped

Month	Number of fruit flies trapped		
	Male	Female	Total
September	7500	7	7507
October	7795	8	7803
November	6180	5	6185
December	6261	9	6270

Table 2
Effect of addition of mango essence to methyl eugenol on attracting fruit fly

Treatments		Average number of fruit flies trapped (mean ± SD)			
		September	**October**	**November**	**December**
A	Methyl eugenol 0.2 mL	190,25 ± 60,85 ab	390,50 ± 102,93 ab	204,00 ± 87,67 b	125,25 ± 42,31 ab
B	Methyl eugenol 0,2 mL + mango essence 0,2 mL	300,75 ± 73,29 bc	268,00 ± 109,24 ab	217,25 ± 61,69 b	283,75 ± 136,43 abc
C	Methyl eugenol 0,2 mL + mango essence 0,4 mL	306,75 ± 100,61 bc	350,25 ± 208,13 ab	340,50 ± 134,77 bc	196,25 ± 59,04 ab
D	Methyl eugenol 0,2 mL + mango essence 0,6 mL	494,25 ± 198,45 cd	448,50 ± 583,35 b	347,25 ± 186,02 bc	545,25 ± 454,82 c
E	Methyl eugenol 0,2 mL mango essence 0,8 mL	584,5 ± 179,91 d	493,25 ± 264,34 b	437,00 ± 214,64 c	415,75 ± 152,65 bc
F	mango essence 0,2mL	0,25 ± 0,5 a	0,00 ± 0 a	0,25 ± 0,50 a	1,50 ± 0,58 a

Means followed by different letters in a column are significantly different ($P < 0.05$, DMRT)

The volatile fruit essence acts as an early trigger of interest in the fruit fly coming into the trap. According to Cosse et al[21], mediterranean fruit fly was attracted to volatile compounds such as ß-pinene, ethyl octanoate, and ß-caryophyllene contained in mango fruits, while the Mexican fly, *Anastrepha ludens* L. was interested in volatile ethyl octanoate compounds[22]. Rattanapun et al[17] reported that female *B. dorsalis* were more attracted to mature mango aroma than to immature mangoes because of the stronger volatile compounds released.

The addition of mango essence on Methyl eugenol was expected to attract female fruit flies into the trap. However, the observation of fruit fly flies during the experiment showed that the interest of female fruit fly was very low. The low interest of female fruit flies may be due to the concentrations of mango essence used in this experiment, so it was difficult to attract the female fruit flies. In addition, durability of fruit essence in nature is not as strong as methyl eugenol. However, the addition of mango essence in methyl eugenol could increase the number of fruit flies trapped during the experiment.

In nature, the application of methyl eugenol traps can reduce the population density of male fruit flies, because it will disrupt the fruit fly behavior in mating so that the occurrence of mating and the number of new individuals produced will be reduced[15]. According to Mahr[23], fruit essence can disrupt the behavior of mating insects caused by the addition of quantity or quality of semi chemical compounds in an area so that supposed addition of essence will cover the male sex pheromone and female fruit flies will get closer to fruit essence than male fruit flies.

The influence of host availability. Biotic factor such as availability of host is a factor that is very influential on the number of fruit fly trapped. The high number of fruit flies trapped in September and October 2016 was due to the production of mango ranges from 20 to 50 kg per tree in these month (table 3). In these months, there was also a fruit maturation phase where there would be an increase in the number of fruit flies because this phase was ideal for laying eggs. The number of fruit flies trapped was lower in November and December than previous months.

Although the availability of fruits was more than 50 kg per tree, farmers sanitized the field by collecting fruits that were attacked by fruit flies in November and harvested mangoes in December. The high density of the fruit fly population in the mango field was directly proportional to the availability of mangoes in the field. Patty[24] found that the greater availability of host in the field, the greater is the potential for fruit fly development. Research conducted by Siwi[25] showed a positive correlation between the level of fruit availability and fruit maturity with the number of fruit flies in the field.

Table 3
The host availability

Month	Number of fruit flies trapped	Production of Mangoes
September	7507	20 kg – 50 kg
October	7803	20 kg – 50 kg
November	6185	> 50 kg
December	6270	> 50 kg

The influence of abiotic factor. Data of temperature, mean rainfall, and mean number of rainy days in Majalengka Regency from September to December 2016 were shown on table 4. The data obtained tended to be stable except the mean rainfall in September which was lower than in other months. The significance values of temperature, mean rainfall, and mean number of rainy days were 0,996, 0,603, and 0,558 respectively (table 5). It showed that based on the

correlation test of regression to these climatic factors, they had no significant correlation to the number of fruit flies.

It was proven based on table 5. Temperature during the study was the optimum temperature for fruit fly development, so it did not affect significantly. *B. dorsalis* complex can live and grow optimally at temperature range 25ºC - 30ºC.[10] The development of fruit flies will be hampered when the temperature is out of the optimum conditions[26]. Other abiotic factors, the high rainfall and number of rainy days may reduce the percentage of successful pupa formed into adult fruit flies. Rain that occurs in the morning or afternoon, the volume and frequency of rainfall, and the number of high rainy days can inhibit the activity of fruit flies[27].

Identification of Fruit Flies. Identification of fruit flies was conducted by observing the fruit fly using a microscope and Delta-intkey software. The results showed that there were three species caught: *B. dorsalis*, *B. carambolae*, and *B. umbrosa* from 100 samples of fruit flies observed (Table 6). *B. dorsalis* had the highest proportion compared to *B. carambola*e and *B. umbrosa*. Kardinan[16] found that fruit flies which had high proportion in Indonesia were from the genus *Bactrocera* and one of the most destructive species was *B. dorsalis* Complex. Siwi[25] found that the common *B. dorsalis* complex species in Indonesia were *B. dorsalis* and *B,carambolae*. The difference among these fruit fly species can be seen from the morphological character of the wing and abdomen (table 7).

Conclusion

The addition of mango essence gave positive effect on performance of methyl eugenol in attracting male and female fruit flies. The highest number of fruit flies trapped was from treatments of 0,6 mL and 0,8 mango essences added to 0,2 methyl eugenol with result rates 347-584 fruit flies per trap in a month. The availability of mangoes and farmer activities also had influence in capturing fruit flies. This current result indicated that mango essence can be used to improve the effectiveness of methyl eugenol traps.

Acknowledgement

The authors would like to thank the Ministry of Technology and Higher Education for providing the scheme of RAPID Skid No.393 / UN6_R / PL / 2016.

Table 4
Data of Abiotic Factors in Majalengka Regency and Number of Fruit Flies Trapped

Months	Temperature (ºC)	Mean Rainfall (mm)	Mean number of rainy day (days)	Number of Fruit Flies Trapped
September	27.7	156.1	21	7507
October	27.3	348.9	23	7803
November	27.6	363.0	21	6185
December	27.3	312.5	22	6270

Source: Meteorology, Climatology and Geophysics Agency of Jatiwangi Meteorological Station, Majalengka Regency, 2016.

Table 5
The Relationship between Number of Fruit Flies Trapped and Abiotic Factors

Abiotic factors	Regression equation	df	R^2	Correlation coefficient	P
Temperature	Y = 7380.1 – 15.9 x	3	0.004	-0.004	0.996
Mean rainfall	Y = 7974.6 – 3.493 x	3	0.397	-0.397	0.603
Mean number of rainy days	Y = -1461.6 + 386.5 x	3	0.442	-0.442	0.558

Note: df = degrees of freedom; R^2 = coefficient of determination; P = significance

Table 6
Proportion of fruit fly species in 100 samples

Sex	Proportion (%)		
	B. dorsalis	***B. carambolae***	***B. umbrosa***
Male	83	5	2
Female	8	2	-

Table 7
Morphological character of *B. dorsalis*, *B. carambolae*, and *B. umbrosa*

Part	*B. dorsalis*	*B. carambolae*	*B. umbrosa*
Imago			
Abdomen			
Thorax			
Wing			

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