J. Trop. Agric. and Fd. Sc. 52 (2)(2024): 59 – 65

The effectiveness of commercial pheromone lures for monitoring fall armyworm (FAW) in Malaysia

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Abstract

The fall armyworm (FAW) is an invasive species that has caused a significant loss of commercial crops worldwide. In Malaysia, fall armyworm was reported in September 2019 with 100% incidence on maize plantations in Changlun, Kedah. Following the reports, the Malaysian Agricultural Research and Development Institute (MARDI), Department of Agriculture Malaysia (DOA), and Centre for Agriculture and Bioscience International (CABI) conducted joint research in developing an Integrated Pest management (IPM) program for managing FAW in Malaysia. One of the components of this program was to identify potential commercial pheromone lures for FAW. Three different pheromone formulations imported from India, the USA, and Costa Rica were evaluated in the laboratory olfactometer assay and the field. The lure from the USA attracts more males compared to the lure from Costa Rica in the choice assay. The field study indicated that the lure from Costa Rica attracted significantly more males, followed by USA and India. Therefore, pheromone lures manufactured in Costa Rica have the potential to be implemented in the IPM program for FAW in Malaysia.

Keywords: *Spodoptera frugiperda, IPM, sex pheromone, monitoring, grain corn*

Introduction

Corn (*Zea mays* L.) is one of the important crops in Malaysia. Generally, there are two types of cultivated corn namely, sweet corn and grain corn. Total production of sweet corn in Malaysia steadily increased over time from 31,907 tonnes in 2003 to 68,907 in 2018 on over 10,362 ha of cultivation area (DOA 2020a). However, in 2020 the total production slightly declined to 68,207 t on a cultivation area of 9,810 ha. The cultivation area of grain corn also showed a similar pattern to sweet corn production where area of cultivation increased from 99.3 ha to 123 ha from 2018 to 2019 but then decreased in 2020 to 75.3 ha (DOA 2020b).

One of the factors leading to a decline in hectare planted is the infestation of insect pests. A recent study has reported the outbreak of Fall Armyworm (FAW) *Spodoptera frugiperda* (J. E. Smith) on corn plantations which severely affected corn production in Malaysia. The FAW has been declared by the Food and Agriculture Organization (FAO) as a major invasive pest in the world with great potential threatening global food security (FAO 2017). The infestation of FAW was responsible for the yield losses of 8.3 – 20.6 million mt/year (21 – 53% of production) in 12-maize producing countries in Africa (Huesing et al. 2018). In Malaysia, FAW was first detected in February 2019 from a corn plantation in Chuping Valley, Perlis. Within a year, this pest has been reported in all states throughout Peninsular Malaysia and Sarawak (Jamil et al. 2021). The severe damage of FAW infestation on corn in Malaysia could cause up to 50 – 100% damage with over 246 ha of area affected (FAO 2019).

To overcome the infestation of FAW, farmers have extensively used chemical pesticides. The chemical approach has helped to suppress FAW, however, continuously using chemical pesticides will cause hazards and pose a threat to health, the environment, and natural enemies (Damalas and Eleftherohorinos 2011). Moreover, FAW is also known to rapidly develop resistance against repeatedly used pesticides, which may lead to decreased

Article history
Received: 23.03.24
Accepted: 28.08.24

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efficiency (Yu 1991). Hence, the implementation of IPM for FAW in Malaysia is needed to effectively manage FAW populations in the country. For the development of the FAW IPM programme in Malaysia, several alternatives to insecticide need to be explored which include the use of biological entomopathogens, the application of effective biorational insecticides, conservation of potential native natural enemies and the implementation of sex pheromone for monitoring of FAW in the field.

Pheromone traps are used worldwide as a monitoring tool for the early detection of FAW and as a guideline for pesticide application in the field (Witzgall et al. 2010). The major component of the female sex pheromone reported from the North American population comprised a mixture of (*Z*)-9- tetradecen-1-yl acetate (*Z*9-14: Ac), (*Z*)-7- dodecen-1-yl acetate (*Z*7-12:Ac), (*Z*)-9- dodecen-1-yl acetate (*Z*9-12:Ac), and (*Z*)-11- hexadecen-1-yl acetate (*Z*11-16:Ac) (Tumlinson et al. 1986). However, this composition was also reported to vary between geographical regions (Batista-Pereira et al. 2006) and FAW strains (Groot et al. 2008). There are several factors influencing sex pheromone lure effectiveness in the field including synthetic compound blends, concentration, trap design, and pest density (Witzgall et al. 2010). In recent study, adding new bioactive component known as nonanal extracted from sex pheromone gland of virgin female FAW significantly increased the attraction of male FAW (Saveer et al. 2023).

Currently, several types of commercial pheromones are registered for FAW and have been used in many countries. To immediately manage this invasive species, Malaysia needs to import these pheromones from overseas, including India, Costa Rica, and the USA. However, no information is available on effective lures for monitoring fall armyworm in Malaysia. Therefore, the efficacy of these pheromones needs to be evaluated for their stability and effectiveness for the implementation in Malaysia. This information is crucial for extension agents and farmers to choose the best pheromone for FAW in the field. For that, laboratory and field evaluations were conducted to assess the effectiveness of imported commercial pheromone lures as monitoring tools for FAW in Malaysia.

Materials and method

Study site

Laboratory evaluation was conducted using a four-arm olfactometer located at Quarantine Laboratory, Horticulture Research Centre, MARDI Headquarters, Serdang, Selangor (2°59'19.6"N 101°42'06.4"E), Malaysia. Field evaluation was conducted at MARDI Centre of Excellence (COE), Bachok, Kelantan (5°58'42.5"N 102°25'34.0"E) on two maize varieties GWG 888 and P4546 in 1 ha.

Commercial pheromones

Three commercial pheromones namely P061-Lure (ChemTica Internacional, Costa Rica), FAW Lure (Pest Control Pvt. Ltd., India), and Trécé Pherocon® Fall Armyworm (Trécé Incorporated, Oklahoma, US) were evaluated in the laboratory and field (*Image 1*). The details of the products including the active ingredients, manufacturer, country and type of packaging are listed in *Table 1*.

Study insects

For laboratory evaluation, FAW colonies obtained from Quarantine Laboratory, Horticulture Research Centre were used for choice assays. Initially, 20 FAW larvae were collected from the sweet corn research plot at MARDI Serdang. Larvae were raised and reared in the laboratory for FAW stock culture. Larvae were fed with fresh corn kernels purchased from a local store. Male FAW of the F3 colony were used in the laboratory study. Male FAW were separated at the pupal stage by observing the distance between the genital and anal opening on the last abdominal segment of each pupa. Male FAW has a shorter distance from genital to anal opening compared with female FAW (Russianzi et al. 2021). Male pupae were raised separately inside a pill box until adult emergence. Unmated adult moths aged 24 – 48 hours were tested in this study.

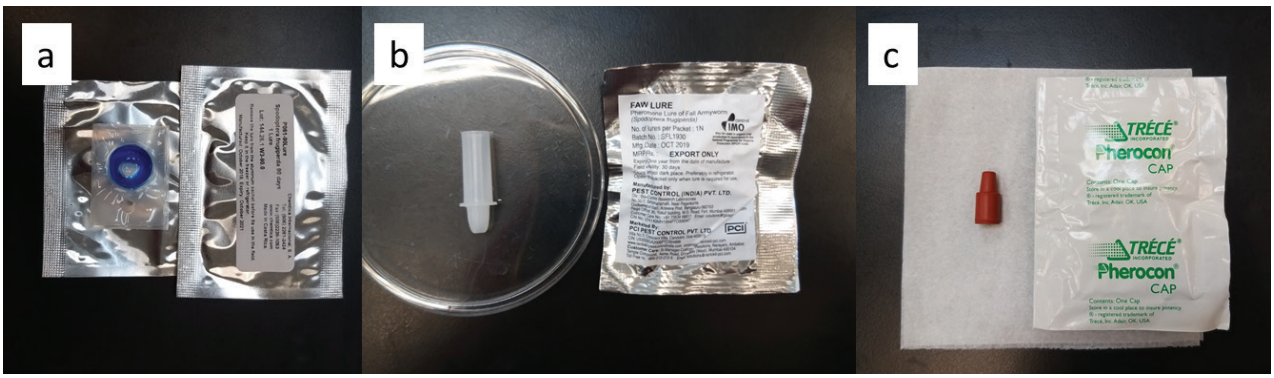

Image 1. Commercial pheromone lures; a) P061-Lure (ChemTica Internacional, Costa Rica) and b) FAW Lure (Pest Control Pvt. Ltd., India) and c) Trécé Pherocon® Fall Armyworm (Trécé Incorporated Oklahoma, US)

Table 1. List of commercial pheromone lures for fall armyworm (FAW) male adult

Name of product	Active ingredients	Manufacturer, country	Packaging
a) P061-Lure	Z7-dodecenyl acetate, Z11-hexadecenyl acetate Z9-tetradecenyl acetate	ChemTica Internacional, S.A. Costa Rica	White plastic bubble
b) FAWLure	Z7-dodecenyl acetate, Z11-hexadecenyl acetate Z9-tetradecenyl acetate	Gaiagen Technologies Private Limited formerly Pest Control Pvt. Ltd., India	Linear low-density polyethylene (LLDPE) vial
c) Trécé Pherocon®	Z7-dodecenyl acetate, Z11-hexadecenyl acetate Z9-tetradecenyl acetate	Trécé Incorporated Oklahoma, US	Rubber Septa

The attractiveness of pheromone lure in the laboratory

Evaluation of three pheromone lures was conducted using a four-arm olfactometer for choice assay. Each end of the olfactometer was provided with one fan powered by a portable power bank of 10000 mAh (Aukey PB-XD12). Both assays were conducted from 20:00 to 05:00 (GMT +8) under dim light.

In this study, each pheromone lure was placed at the end of each arm. One end of the olfactometer arm was left empty as a control. One male FAW was released into the main chamber, and the choice made by each male FAW was recorded every 5 minutes until 30 minutes of observation (*Image 2*). This step was repeated with 20 males FAW for 10 replications. Total of 200 males were tested in this study, only 125 were responsive and made a choice within 30 minutes of observation. The olfactometer was rotated 180° after five males tested to reduce the positional effect. Each chamber was cleaned with 70% alcohol, and the fan on each arm was allowed to blow for 5 minutes before the initiation of the next replicate. In this assay, male adults were subjected to a single test, while unresponsive male adults were not considered for the analysis.

Field evaluation of commercial pheromone

Field evaluations were conducted in two planting seasons (Image 3). In season 1, two pheromone lures, P061-Lure (ChemTica Internacional, Costa Rica) and FAW Lure (Pest Control Pvt. Ltd., India) were installed in a Delta trap using replicated 3 x 3 Latin square design consisting of 3 treatments (rows) by 3 positions (columns) in 18 consecutive weeks (replications). Trap with no lures were used as control. Delta traps layered with white sticky sheet were spaced 60 m apart in a triangular arrangement and hung 1.5 m above the ground. Replacement of white sticky sheets and trap rotation (clockwise) were made weekly.

In season 2, Trécé Pherocon® Fall Armyworm manufactured in the USA was added as the fourth treatment. All four treatments were arranged in a replicated Latin square design (4 treatments by 4 positions) with a similar setting as in season 1 for 12 consecutive weeks (three replications).

In both seasons, pheromone lures were replaced after 4 weeks of exposure in the field. Used sticky sheets were collected weekly and male FAW trapped on the sticky sheets were recorded.

Image 2. Experimental arena for choice assay using a four-arm olfactometer

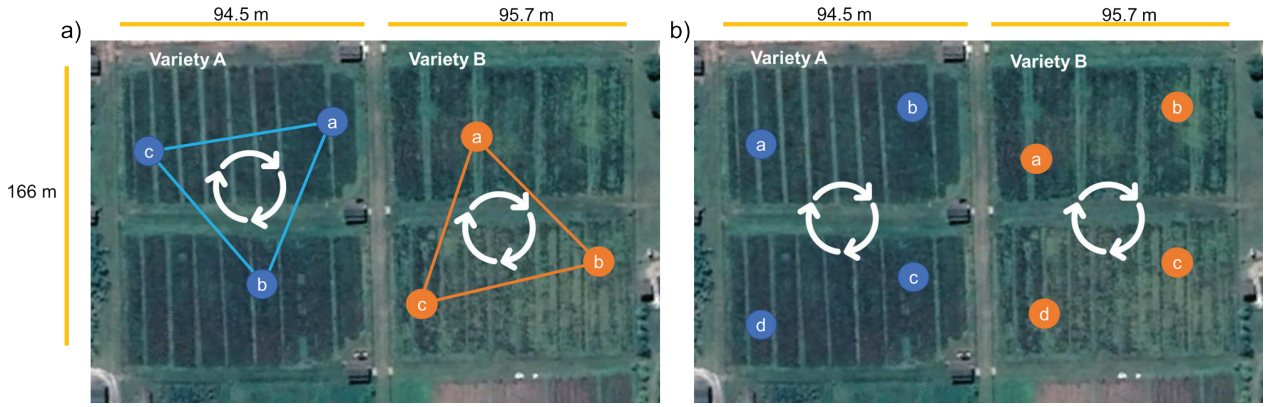

Image 3. Latin square design for (a) season 1 (3 treatments by 3 positions) and (b) season 2 (4 treatments by 4 positions) on different grain corn varieties, (variety A: GWG 888; variety B: P4546) at MARDI Bachok, Kelantan

Statistical analysis

In the choice assay, the average number of m ales that made a choice for each treatment was calculated and analysed using one-way ANOVA (analysis of variance). In the field study, the number of male FAW captured for each treatment (row) on each position (column) was counted weekly. The data were then analysed using a two-way ANOVA. In both studies, means were compared using Tukey's Studentized Range (HSD) Test at a significance level of $P < 0.05$.

Results and discussion

The attractiveness of pheromone lure in the laboratory

In the choice assay, the Trécé Pherocon® FAW lure attracted numerically more FAW than the FAWLure and statistically more than P061-Lure. There was no statistical difference between the FAWLure and the P061-Lure (F=17.20; df=3,12; *P*=0.0001) (*Table 2*). The result of this trial suggested that all lures could be used to attract male FAW in Malaysia. Therefore, all lures were further evaluated in the field to assess the effectiveness in attracting wild FAW in Malaysia.

Table 2. Mean ± SE number of male FAW attracted to different type of pheromone lure in choice assay in the laboratory

Type of pheromone	Choice assay
T1: Control	2.43 ± 0.72 b
T2: P061-Lure	2.57 ± 0.27 b
T3: FAWLure	4.07 ± 0.62 ab
T4: Trécé Pherocon®	4.86 ± 0.48 a
	F=4.66; df=3,52; *P*=0.0059

Mean in the same column marked by different letters are significantly different by Tukey's honestly significant difference test (Tukey's HSD) at $P < 0.05$

Field evaluation of commercial pheromone

Overall, the P061-Lure has shown greater attractiveness to FAW compared with FAWLure and Trécé Pherocon® under the temperature range 25 – 30 °C and 75 – 96% RH.

In season 1, the P061-Lure captured significantly more (3.5x) male FAW then the FAWLure (F=67.19; df=2,42; *P*<0.0001) (*Table 3*). The attractiveness of the FAWLure declined after 2 weeks exposure in the field, whereas the P061-Lure consistently attracting male FAW up to 4 weeks (*Figure 1*).

Similarly, the P061-Lure (7.71 ± 1.04) recorded significantly higher numbers of FAW in season 2 than Trécé Pherocon® (4.71 ± 0.91) and FAWLure (2.88 ± 0.86) (F=5.79; df=3,88; *P*<0.0001). The P061-Lure has been reported from Argentina, Mexico, and Brazil to have higher effectiveness in attracting FAW than Trécé Pherocon® (Malo et al. 2001; Cruz et al. 2012). Although Trécé Pherocon® has lower number of FAW trapped compared to P061-Lure, the durability of Trécé Pherocon® in attracting FAW in the field was similar to P061-Lure (*Figure 2*). There were no significant differences recorded for different maize varieties and trap positioning in both seasons.

Koffi et al. (2021) suggested that the composition of pheromone lure blends influenced the number of FAW captured. In this study, all three lures have a similar component mixture which includes (*Z*)-9- tetradecen-1-yl acetate (*Z*9-14: Ac), (*Z*)-7- dodecen-1-yl acetate (Z7-12:Ac) and (*Z*)-11- hexadecen-1-yl acetate (*Z*11-16:Ac). However, the ratio of each component might vary depending on their manufacturer. The reported mixture of chemical blends was insufficient to explain the differences in lure performance in the field. However, these differences could be influenced by the type of dispenser used for the pheromone. The P061-Lure was in a form of blue liquid loaded in bubble capsules, while FAWLure was infused in Linear low-density polyethylene (LLDPE) vial and Trécé Pherocon® was in red rubber septa.

Table 3. Mean ± SE number of male FAW adults recorded weekly in the delta traps installed with different types of pheromone lures and a control trap (no lure) in the field study of season 1 and season 2

Type of pheromones	Season 1		Season 2	
	PLOT A	**PLOT B**	**PLOT A**	**PLOT B**
T1: Control	0.00 ± 0.00 b	0.00 ± 0.00 b	0.33 ± 0.19 c	0.75 ± 0.33 c
T2: P061-Lure	4.87 ± 0.76 a	7.27 ± 1.10 a	8.17 ± 1.57 a	7.25 ± 1.41 a
T3: FAWLure	1.93 ± 0.64 b	1.47 ± 0.51 b	2.92 ± 1.03 bc	2.83 ± 1.42 bc
T4: Trécé Pherocon®	Not included in the treatment		5.33 ± 1.44 ab	4.08 ± 1.16 b
	F=15.71 df=2,10; *P*=0.0008	F=24.02; df=2,10; *P*=0.0002	F=12.36; df=3,18; *P*=0.0001	F=5.79; df=3,88; *P* <0.0001

Mean in the same column marked by different letters are significantly different by Tukey's honestly significant difference test (Tukey's HSD) at *P* <0.05

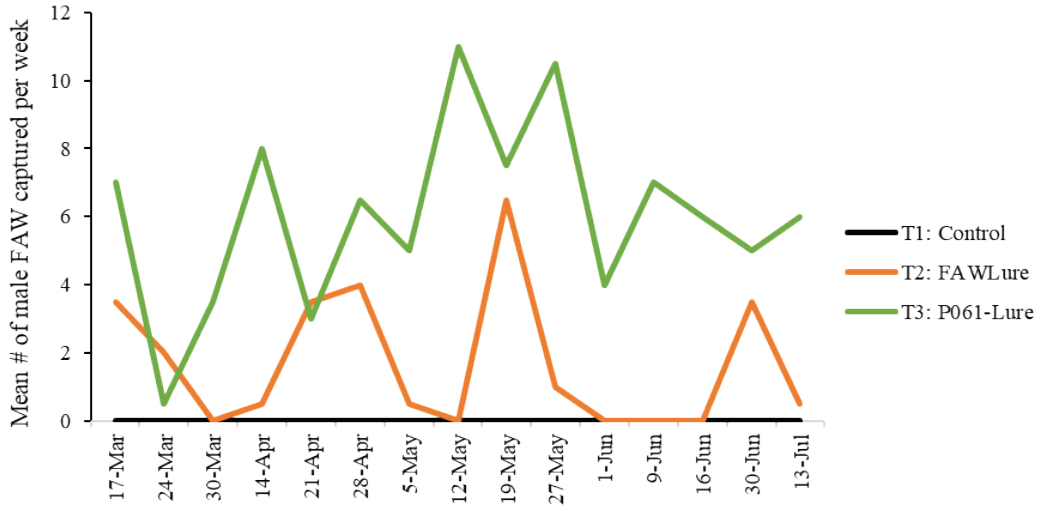

Figure 1. Weekly count of FAW population in season 1 over a period of 18 weeks

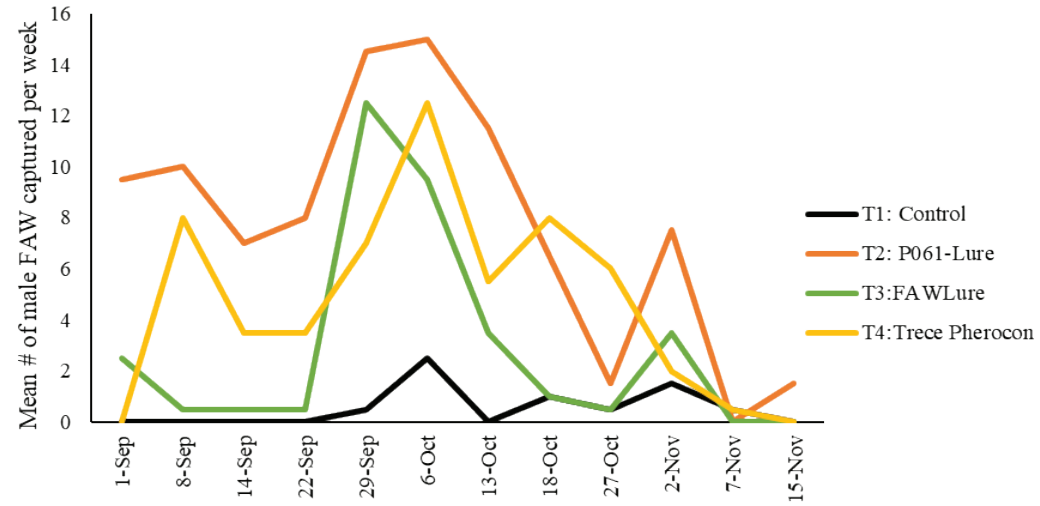

Figure 2. Weekly count of FAW population in season 2 over a period of 12 weeks

The bubble capsule technology was invented by Axel Meisen (University of British Columbia, Canada) and was made from a semipermeable membrane blister package that allowed for efficient slow release of pheromone blends and long-lasting (Ross 2021). Our study reported that pheromone P061-Lure consistently captured male FAW for four weeks (*Figure 1*) which was similar to the previous study reported by Holsten (2022).

The number of FAW captured in this study was relatively similar to the study by Cruz-Esteban et al. (2022), which recorded 10 males/trap/night when a delta trap was used in the trapping system. Although the same study reported more males captured when different trap designs were used, the delta trap was considered the best option for our study. This is because the large sticky surface reduced damage on FAW wing scales, allowing for accurate identification of trapped FAW. Cruz-Esteban et al. (2022) concluded that trap design plays an important role in influencing the effectiveness of a trapping system using a sex pheromone lure. Additionally, Saveer et al. (2023) showed that the number of male catches increased by 53–135% with the addition of nonanal to the commercial pheromone. Therefore, future studies should consider adding nonanal and optimising trap designs to maximise the number of male FAW captured per night.

Conclusion

The laboratory studies evaluating the attractiveness of all three lures on male FAW suggested that all three lures effectively attract male FAW. However, field evaluation on all lures using the delta trap, showed that the pheromone from Costa Rica consistently captured a higher number of male FAW over two planting seasons. This finding suggests that lure the P061-Lure has the potential to be included in the monitoring strategy for the FAW IPM programme in Malaysia. Therefore, future studies on trap design and placement as well as lure longevity should be conducted to obtain an effective pheromone trapping system for FAW in Malaysia.

Acknowledgement

This study was conducted as part of a collaboration project between MARDI and CABI to develop an Integrated Pest Management (IPM) Programme for Fall Armyworm (FAW) in Malaysia. The authors would like to acknowledge Nur Khairani Abu Bakar, Mohd Saidi Zakari, Nurul Farhani Mohamed and the staffs at MARDI Bachok, Kelantan, for their technical assistance while conducting this research.

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