# Using Pheromone Mass Trapping Within IPM Programmes For Controlling The Red Palm Weevil, *Rhyncophorus ferrugineus* Olivier

**Sewify, G. H.[1]; Belal, M. H.[1] and Saeed, M. Q.[2]**
[1] Department of Economic Entomology and Pesticide, Faculty of Agriculture, Cairo University.
[2] Department of plant Protection, Ministry of Environment and Agriculture, Doha-Qatar, P.O.Box 10619.

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## ABSTRACT

A field experiment was conducted to evaluate the integrated effect of baited aggregation pheromone traps and entomopathogenic fungus *Beauveria bassiana* or insecticide for controlling the red palm weevil (RPW),*Rhyncophorus ferrugineus* Olivier (Coleoptera: Rhynchophoridae) through 2008-2009. Results indicated that the intensity of captured weevils were decreased as a result of using combination of baited pheromone traps and fungus or insecticide compared with using baited pheromone trap only. Statistical analysis revealed significant differences between the mean numbers of captured adults in treated areas with baited pheromone traps alone and combination of traps and fungus or insecticide whereas the total mean numbers were 17.04,4.93 and 8.02 adults/3traps, respectively. The total mean reduction of RPW population caused by mass trapping and fungus *B. bassiana* or insecticide were 61.40% and 40.16% , respectively. Considerable reduction of infested palm tree numbers was noticed in treated areas with combination compared with untreated areas. There were significant differences among different field trials and untreated areas on mean numbers of infested palms. They were lower (0.77 and 0.82 palms) when used combination of baited pheromone traps plus fungus and baited pheromone traps plus insecticide, respectively and higher (2.03 and 2.15 palms) when used mass trapping or insecticide alone. However the untreated areas recorded highest infested palm trees whereas mean number was 3.73 palms.

***Key word: Rhyncophorus ferrugineus , Baited pheromone traps, Beauveria bassiana, IPM, Date palm***

## INTRODUCTION

The red palm weevil (RPW) *Rhyncophorus ferrugineus* Olivier (Coleoptera: Rhynchophoridae) is a damaging pest of palms distributed in diverse ecological habitats and is reported to attack 17 species of palms in nearly 50% of the date palm-growing countries (Faleiro, 2006). Although, the native home of the RPW is South Asia where it attacks coconut, *Cocus nucifera*, during the last two decades it has invaded countries in the Middle East and Europe where it has become a serious threat to the cultivation of date palm, *Phoenix dactylifera.* It invaded Egypt in 1992 (Saleh, 1992) where it is now causing sever damage to date palm. RPW is a concealed tissue borer difficult to detect. Infested palms detected in the early stage of attack are easy to control with insecticide. However, palms in the late stage of attack do not respond to chemical treatments and often die. Usually palms below the age of 20 years are preferred by the pest. Current control methods include surveys to identify infested palms, removal, burning or burial of heavily infested trees, injection insecticide into infested palms and spraying of insecticide on all palms in infested areas. Burning of palm trunks is often incomplete, so some larvae and pupae survive and complete development. Delay between detection and destruction of an infested tree permits emergence and migration of adult weevils prior to destruction. Transporting infested trees and off-shoots for burning introduces the weevil to new areas (Hallett *et. al.*, 1999). The most effective and environmentally sound method is the food-baited pheromone trap (Abraham *et. al.*, 1998; *Faleiro et.al.,*1998). These traps have been successfully used as mass trapping devices to manage the red palm weevil on date palm farms in the Middle East. It has been reported that using aggregation pheromone traps were very effective in reducing the adults the red palm weevil and curtailing population growth in the field (*Abraham et. al.,* 1999, 2000; Faleiro, 2000). The entomopathogenic fungus *Beauveria bassiana* (Balsamo)Vuillemen has been successfully used as a bio-control agent for the management of a number of Coleopteran insects including the colorado potato beetle , *Leptinatarsa decemlineata* Say (Anderson 1988), Lesser mealworm , *Alphitobius diaperinus* Panzer (Geden *et. al*, 1998 ) , blister beetle , *Lytta nuttalii* Say (Miranpuri *et. al.*, 1994) and maize weevil, *Sitophilus zeamais* Motschulsky (Meikle *et. al*, 2001) In Egypt , it has been found infecting the pink borer , *Sesamia cretica* (lederer), European corn borer , *Ostrinia nubilalis* (Hibner), whitefly *Bemisia tabaci* and weevil *Hpyera brunneipennis* , Boheman (Ezz , 2004, Sewify, 1999 , El-Sufty and Hrer, 1989). All *R . ferrugineus* stages showed different response susceptibilities to *B . bassiana* infection under laboratory conditions (Aldossary *et. al.*, 2009 and El – Sufty *et.al.*,2009). These are especially important for controlling cryptic insects such as red palm weevil which are not accessible to its natural enemies. The use of entomopathogenic fungus *B. bassiana* against red palm weevil has been reviewed by Sewify and Fouad (2006) and Sewify *et. al.*(2009).

The aim of the present work was to evaluate the using of combination of mass trapping aggregation pheromone and fungus *B. bassiana* or insecticide within IPM programme for controlling the red palm weevil.

## MATERIALS AND METHODS

**Study sites**

*R. ferrugineus* inhabit large scale palm date belt extended allover Ismailia Governorate (100km far from Cairo) thus covering an area of 450,000 Feddan, in which date palm trees were extensively cultivated. The experimental area was chosen at Kasassin region through 2008-2009.

**Using mass trapping within IPM programmes for controlling RPW:**

A field experiment was conducted through 2008-2009 to evaluate the integrated effect of baited aggregation pheromone traps and biological control by using entomopathogenic fungus *B. bassiana* or insecticide for controlling RPW. Five palm farms containing 430, 470 ,450, 410 and 700 palm trees were selected, each around 25 feddans, with a distance of 2 Km. between each other farm. The palm farms were similar in variety of plant, age ,size and climate. Three replicates were conducted for every treatment (5 feddans). Each farm received one of the following five treatments. (A) using pheromone-baited traps only (3traps/5feddans); (B) using pheromone-baited traps (3traps/5faddens) and entomopathogenic fungus *B. bassiana* at concentration of $5x10^7$ spores/ml; (C) application of insecticide Tafaban 48% (Chlorpyifos) at recommended dose (3cm/L.). (D) using pheromone baited traps (3traps/5faddens) and insecticide Tafaban 48% (Chlorpyifos) at recommended dose (3cm/L.). (E) untreated control. Farms treated with entomopathogenic fungus or insecticide received two applications, on the Mid March and the second on the first week of September 2008. Trials lasted 52 weeks through 2008-2009 and damaged trees during trapping period and captured weevils were recorded weekly.

**Trap Design**

Pheromone traps were fabricated using a 10 L polypropylene bucket with four rectangular (3x8 cm) windows cut equidistantly below the upper rim of the bucket. The distance between each window and the bottom of the bucket was 16 cm. The bucket was covered with a lid that had three windows similar to the ones on its sides. The upper surface of a lid had a small handle to opening the trap and the lower side had a small knob on which a wire was fixed to hold the pheromone and kairomone dispensers (Al-Saoud *et.al*2010) The commercially used pheromone "Ferrugineol" is a synthetic pheromone lures.It is a mixture of 4 – methyl – 5 nanol and 4 – methyl – 5 nanone (9:1) imported from Chem. Tica Natural, Costa Rica and was used for the present field trials. Pheromone was hanged underside the trap lid. The pheromone releases its active chemicals through a plastic membrane (0.3 and 1.0 mg/day) from 400 and 1500 N/tube, respectively under laboratory conditions of 27C and 50% R.H.. Ethyl acetate, kiromone was used as a synergist to activate the potent ability of releasing pheromone. Ethyl acetate bottles were hanged from the underside surface of the trap lid releasing chemicals through a fine plastic tube (as 100 to 128 mg/d). The traps were supplemented with a fermenting mixture of sugarcane pieces and sugarcane molasses. Some soap was mixed with poisoned water in thebucket. Servicing of traps was conducted to keep them wet and food freshen every 2 weeks. Ethyl acetate Kiromone and pheromone capsules within each trap were changed by another fresh ones monthly.

**Fungal inoculum:**

The entomopathogenic fungus *B.bassiana* used in the experiments was originally isolated from red palm weevil *R.ferrugineus* found in Ismailia Governorate in Egypt .This fungus was grown on autoclaved Sabouraud and dextrose yeast agar (SDAY), containing 1% peptone ,0.2%yeast extract ,4%dextrose and 1.5%.agar in distilled water and incubated for two weeks at 26C°+1

**Conidiospores mass production:**

*B .bassiana* aerial conidia were produced using biphasic culture system (Bradley *et. al.*, 2002, Leland *et al.*,2005). Flasks(100 ml) of liquid biomalt medium (25 g biomalt and 2 g yeast extract),were incubated for 3-4 days at 25C° The liquid cultures were then used to inoculate autoclaved white rice in sterilized plastic bags. The rice was first soaked in water for 12 hours and autoclaved for 30 min in autoclaveable plastic bags (60x80 cm) in the ratio of 2 kg rice / bag at 103 k pa for 20 min. After cooling, three to four day the old liquid culture of *B. bassiana* was mixed by hand with the substrate ,under aseptic condition , at the ratio of 50ml /bag . An absorbent cotton plug rolled around tube (15cm long) had been used to plug each bag. The bags were connected with the source of filtered air through air valves .Solid substrate fermentation was kept for 11 days at 26C° Culture was observed daily and crumbled by hand within bags to prevent binding of the substrate Whole culture was then transferred to wooden screen shelves where culture was dried for 7days at 28C°. Conidia were harvested by mechanical sieving.

**Fungal and insecticide spraying**

All palms (2 - 10 years– old) were numbered. The damage by the red palm weevil at the selected sites was severe and had already led to the loss of several palms. The conidiospores suspension was used in a final concentration of 5 x $10^7$ spores / ml formulated with sun flower oil. The fungus and insecticide was applied by using high pressure motor sprayer (400L) to cover the trunk of palm tree. The Spray nozzle was directed at the apical part of the trunk and at leaf axils. Spraying continued to the point of run–off, until all trunk and leaf axel were fully saturated, using 8 – 10 L of the spray suspension per tree.

Weevils were collected and counted weekly .The efficiency of such treatments was based on the reduction in RPW population captured by traps according to formulate of Henderson and Tilton (1952).

**Statistical analysis**

The data were subjected to ANOVA using the PROC GLM procedure and the means were compared by carrying out the Least Significant Difference (LSD) procedure of the SAS statistical package (SAS, 2001).

## RESULTS AND DISCUSSION

**1. Effect of combinatio n of baited pheromone traps and fungus *B. bassiana* or insecticide on population density of RPW.**

The illustrated data in (Fig.1) indicated that the intensity of captured weevils decreased as a result of used combination of baited pheromone traps and fungus or insecticide compared with those recorded using only baited pheromone trap. Conversely to the site used pheromone traps only, the RPW population density in site treated with combination of baited pheromone traps and fungus *B. bassiana* decreased from 20.3adults/3traps in March to 12.7 adults/3traps in April and still decreased until end of August and then increased in September and October then decreased rapidly up to 1.3 adults/3traps in March 2009.

The same trend was noticed in the site received a combination of baited pheromone traps and insecticide, whereas the mean number of captured weevil decreased from 20.3 adults/3traps in March 2008 to 12.7 adults/3traps in April and decreased steadily up to 3.3 adults/3traps in march 2009. Statistical analysis revealed significant differences between the mean numbers of captured adults in treated areas with baited pheromone only and a combination of traps and fungus or insecticide whereas the total mean trapped numbers were 17.04,4.93 and 8.02 adults/3traps, respectively (Fig.2).

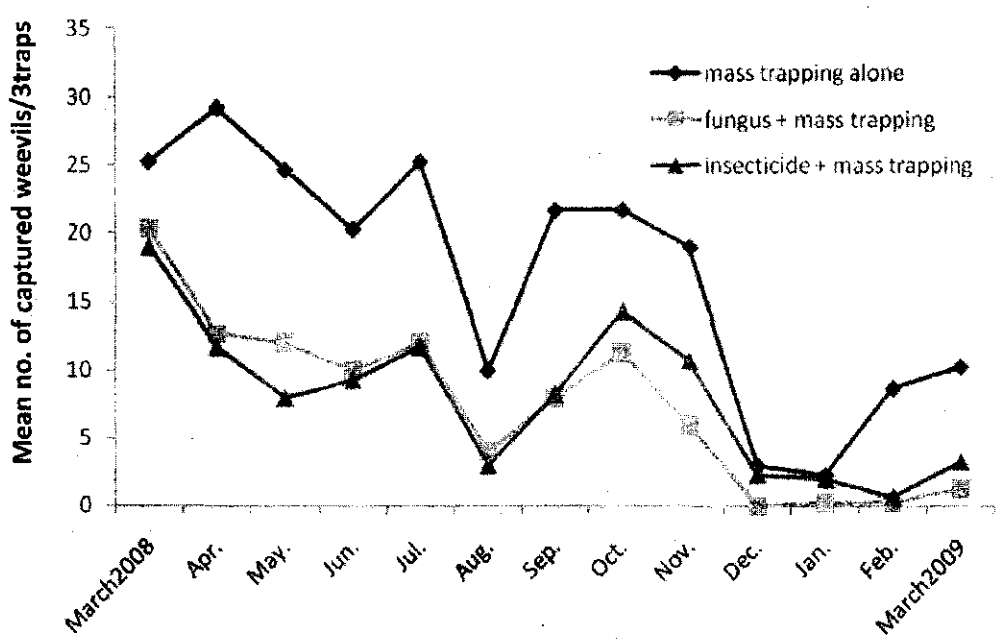


**Fig. 1: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on monthly mean numbers of RPW during 2008-2009.**

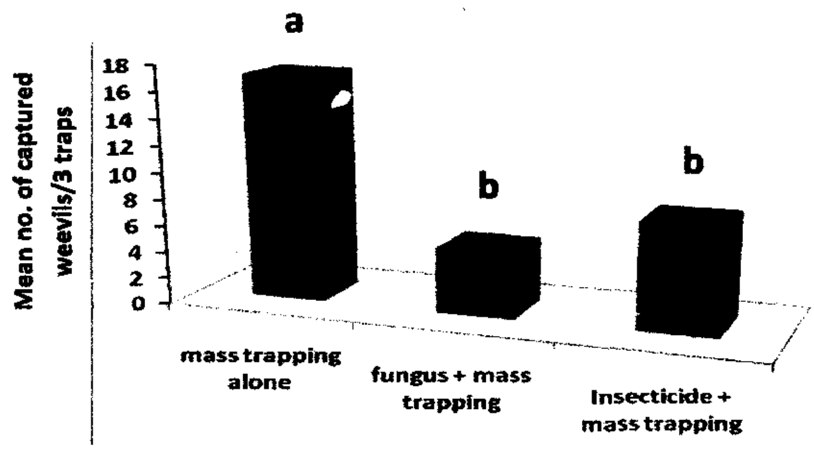


**Fig. 2: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on total mean numbers of captured weevils during 2008-2009. Columns labeled with the same letter are not significantly different (p>0.05).**

**2. Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide in reduction of captured weevil of RPW**

The illustrated data in Fig. (3) showed the reduction of captured weevils of RPW induced by using a combination of baited pheromone traps and fungus *B. bassiana* or insecticide. In April (2008) recorded reduction was 46.20% and 46.97% for the two applications, respectively. The reduction rate of captured weevils increased again in August 2008 to reach 60.66% and 68.42 for both applications, respectively. The application of combination of baited pheromone traps and insecticide decreased rapidly up to.15% and 0% in November and December, respectively while the application of baited pheromone traps and fungus increased the reduction rate to reach 60.6% and 100% at the same months, respectively. This indicates actually, establishment of fungal efficacy in RPW population. Generally the total mean reduction of RPW population caused by mass trapping and fungus *B. bassiana* or insecticide were 61.40% and 40.16%, respectively.

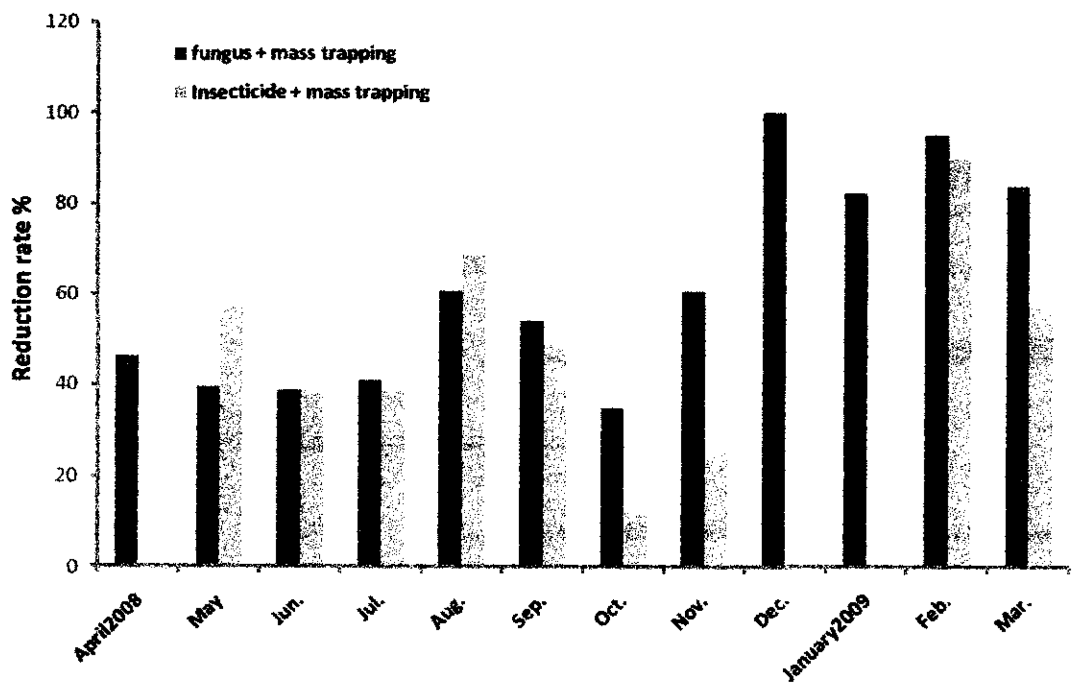


**Fig. 3: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on reduction rate of captured weevil of RPW during 2008-2009.**

**3. Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on infested palm trees.**

Considerable reduction of infested palm trees numbers was noticed in treated site<sup></sup> with combination of baited pheromone traps and fungus *B. bassiana* or insecticide compared with untreated ones. Fig. (4) shows the effect of different treatments on monthly mean numbers of infested palm trees compared with untreated control through 2008-2009. There were significant differences on the impact of different field trials and untreated areas on total mean numbers of infested palm. They were lower (0.77 and 0.82 palms) when used combination of baited pheromone traps + fungus and baited pheromone traps + insecticide, respectively versus higher (2.03 and 2.15 palms) when used mass trapping or insecticide only. However, the untreated sites recorded highest infested palm trees whereas mean number was 3.73 palms (Fig.5).The results revealed that there were no significant differences between the effect of combination of baited pheromone traps + fungus and baited pheromone traps + insecticide on reducing infested palm trees. In order to compare the efficacy of different treatments for the control of RPW, the percentage of reduction was converted to control rates as shown in Fig. (6).

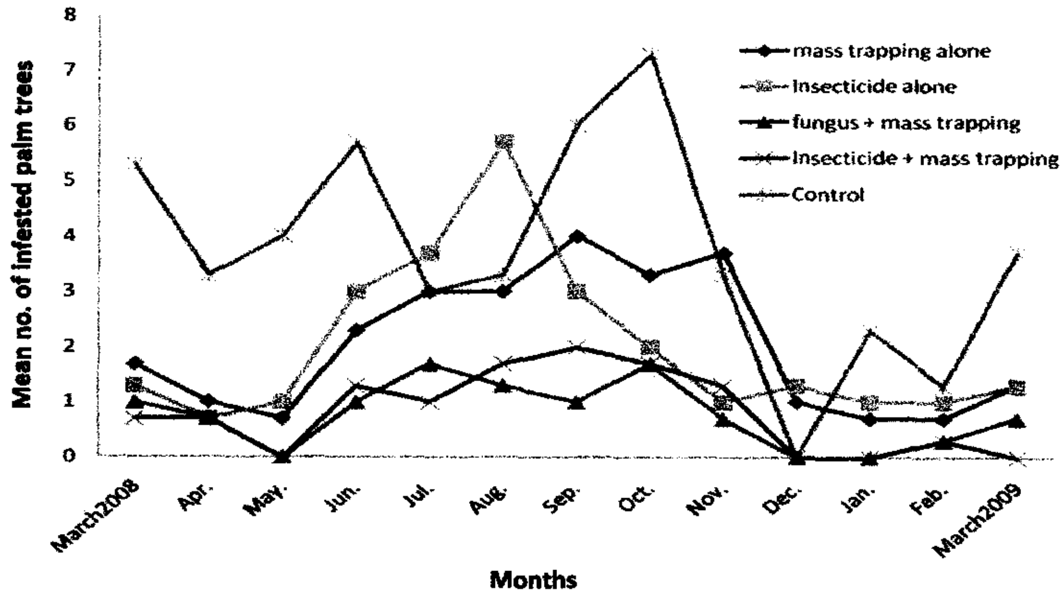


**Fig. 4: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on monthly mean numbers of infested palm trees during 2008-2009.**

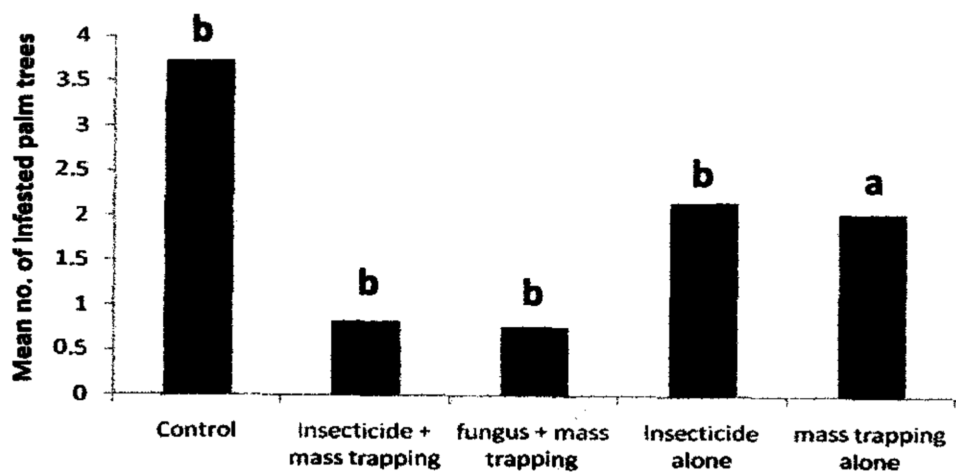


**Fig. 5: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on total mean numbers of infested palm trees during 2008-2009. Columns labeled with the same letter are not significantly different (p>0.05).**

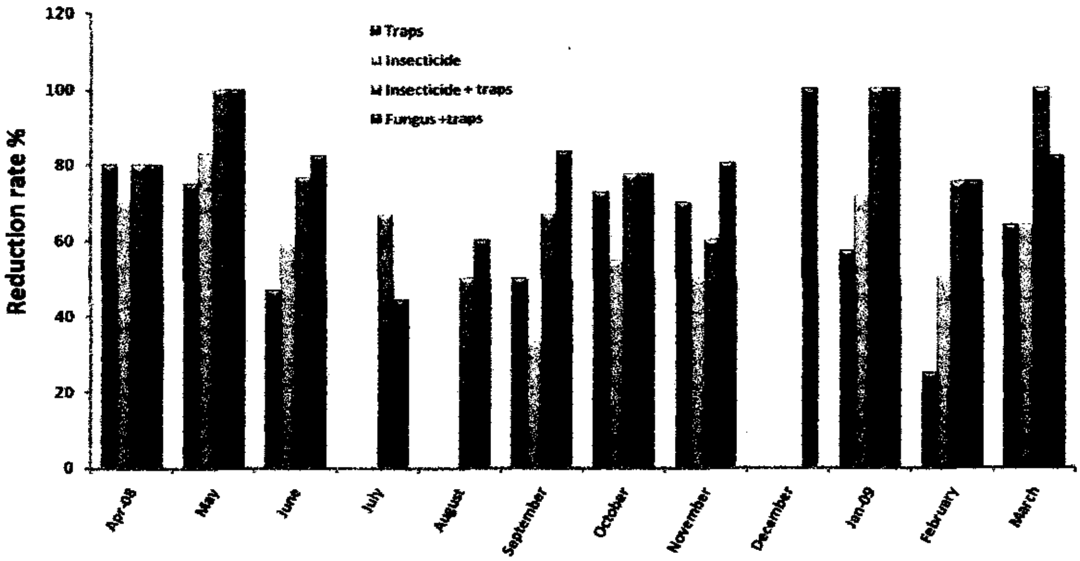


**Fig. 6: Effect of combination of baited pheromone traps and fungus *B. bassiana* or insecticide on reduction rate of infested palm trees.**

Generally the total mean reduction of infested palm trees caused by using baited phermone mass trapping alone and combination of baited phermone mass trapping with fungus *B. bassiana* or insecticide were 44.59% , 80.35% and 71.02%, respectively

The results confirmed that field treatment with combination of baited aggregation pheromone and entomopathogenic fungus *B. bassiana* successfully reduced the field RPW population and infested palm trees.

Mass trapping with pheromone-baited traps is considered to be one of the major components of IPM programmes for controlling RPW. The components of the RPW-IPM programme in date palm farms are early detection of infested palms, monitoring insect pests, field sanitation, trapping of adult weevils using baited pheromone traps, preventive and curative, chemical treatments, eradication of severely infested palm trees, biological control by entomopathogenic fungus and pathogenic nematodes and implementation of quarantine measures (Abraham *et.al.,* 1998, Faleiro, 2006 and Sewify,*et al.* 2009). In the present study ,combination of baited pheromone traps (3traps/feddens) and entomopathogenic fungus or insecticide Tafaban 48% decreased significantly the mean numbers of captured adults compared with those recorded by baited pheromone traps alone. The combination induced significant reduction in the capture mean numbers of RPW up to. 61.4 and 40.16% for traps+fubgus and traps+insecticide, through 2008-2009, respectively. Sewify *et al.,* (2009) reported that fungal *B. bassiana* application by dusting conidiospores and/or releasing the fungal contaminated RPW males decreased the population of captured weevils in treated plots compared with untreated ones. This fungus has been proven to be effective control agent for many Coleopteran species, (Miranpuri, *et .al.,* 1992a and 1992b, Miranpuri, Khachatourians, 1994; Athanassiou and Steenberg, 2007 and Aldossary *et.al.,* 2009). Combination of baited pheromone traps and fungus *B. bassiana* or insecticide Tafaban decreased significantly the numbers of infested palm trees in treated sites compared with untreated ones. In the Al-Hassa region of Saudi Arabia, mass trapping of RPW, between 1994 and 1998, maintained infestation levels below 1% in the endemic areas of c. 4000 ha of date plantations, whilerestricting its spread in the relatively pest-free areas (Abraham *et. al.,* 2000). Similarly, in the Al-Qateef region of Saudi Arabia, infestation levels were reduced from 7% in 1993 to 3% in 1997 using RPW-IPM programme in which pheromone trapping played a key role. Weevils captured reduced insect population from four in 1994 to two weevil per trap per week, at the end 1997 (Vidyasagar *et.al.,* 2000a,b). Mass trapping the pest along with other RPW-IPM tactics for 2 years between 1999 and 2001, on 450 ha of date plantations in Israel, resulted in a significantly decreased number of weevils trapped by the end of 2001 (Soroker *et.al.,* 2005). In another study from the UAE, pheromone trapping for 1 year on different farms insecticides-free resulted in reduction of infestation by 71% (Oehlschlager, 2006). Implementation of a pheromone-based RPW-IPM programme in the Sultanate of Oman resulted in reduction of infested date palm by weevils , from 24% in 1998 to only 3% in 2003 (Al-Khatri, 2004).

Results of the present work suggest that IPM depending on trapping of adult weevils and biological control with entomopathogenic fungus

*B. bassiana* besides chemical control might be best offer chance for success.

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## الملخص العربى

# استخدام الاصطياد المكثف للمصائد الفيرمونية في إطار المكافحة المتكاملة لمكافحة حشرة سوسة النخيل الحمراء

جمال حسن سويفى[1] ، محمد حلمى بلال[1] ، محمد قائد سعيد[2]
[1]قسم الحشرات الاقتصادية والمبيدات – كلية الزراعة – جامعة القاهرة
[2]قسم وقاية النبات ،وزارة البيئة والزراعة – الدوحة – قطر

أجريت تجربة حقلية لتقييم التأثير المشترك للمصائد الفيرمونية والمكافحة الحيوية باستخدام الفطر الممرض للحشرات *Beauveria bassiana* والمبيد الحشري تافابان ٤٨% وذلك خلال عامي ٢٠٠٨- ٢٠٠٩. وقد أظهرت النتائج أن استخدام المصائد الفيرمونية مع الفطر الممرض اوالمبيد أدى إلى حدوث نقص معنوي في متوسطات أعداد الحشرات الملتقطة في المناطق المعاملة بالمصائد مع الفطر أو المبيد عن متوسط أعداد الحشرات الملتقطة في المناطق المستخدم بها المصائد الفيرمونية فقط حيث كانت متوسطات أعداد الحشرات الملتقطة في المناطق المستخدم فيها المصائد مع الفطر، والمصائد مع المبيد، والمصائد فقط ٧,٠٤ ، ٤,٩٣ ، ٨,٠٢ حشرة كاملة / ٣ مصيدة على التوالي . وقد أحدثت المعاملة بالمصائد الفيرمونية والفطر الممرض أو المبيد الحشري خفض كلى فى أعداد الحشرات الملتقطة بلغ ٤٠,١٦% و ١٦,٤٠ %على التوالي. كذلك أظهرت النتائج حدوث خفض معنوي فى أعداد النخيل المصاب بحشرة سوسة النخيل الحمراء في المناطق المعاملة بالمصائد الفيرمونية والفطر الممرض أو المبيد الحشري مقارنة بالمناطق غير المعاملة. وتوضح النتائج أن اقل متوسط للنخيل المصاب بحشرة سوسة النخيل الحمراء كان في المعاملات التي استخدم فيها المصائد الفيرمونية مع الفطر أو المبيد (٠,٧٧ و٠,٨٢ نخلة مصابة) بينما كان أعلى متوسط للإصابة في المعاملات التي استخدم فيها المبيد فقط أو المصائد فقط (٢,٠٣و ٢,١٥ نخلة مصابة ) بينما وصل متوسط أعداد النخيل المصاب في المناطق الغير معاملة ٣,٧٣ نخلة مصابة.