

Ramy Aljazzar

**New Bait-Based Techniques for the Control
of the Peach Fruit Fly *Bactrocera zonata*
(Saunders) and the Mediterranean Fruit Fly
Ceratitis capitata (Wiedemann)**

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Dedication

This work is dedicated to

My beloved mother, who has incessantly and tirelessly encouraged me to excel in my academic studies and professional life since my childhood;

My late father and elder brother, who taught me as a young teenager and without a book the skills of critical thinking and deductive and inductive reasoning, which I believe have become part of my character.

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Ramy Aljazzar

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- **Dr. Khaled Djelouah**, Division of Integrated Pest Management (IPM), Mediterranean Agronomic Institute of Bari (MAIB), Italy.

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Chapter 1

Introduction

Apricot, *Prunus armeniaca* (L.) is native of Asia. The tree is small (6–10 m tall), and it has long been cultivated in China, India, Egypt, and Iran. It is now grown in Europe, parts of Africa, and the warmer parts of the New World. This tree is susceptible to frost, so it is grown in warm temperate regions and subtropical regions (Hill, 2008).

According to FAO estimates, the world production of apricots mounted to 3,834,474.7 tons in 2011. In Egypt, the production of apricots was estimated at 96,643 tons in the same year, which constitutes about 2.5 percent of world production (FAOSTAT 2011 estimates). While Egypt is not one of the world's top producers of apricots, the country ranks third globally in terms of highest yields, with a yield of 154,703.06 hectograms per hectare (FAOSTAT 2011 estimates).

The San José scale *Quadraspidiotus perniciosus* (Comst.), peach twig borer *Anarsia lineatella* (Zell.), peach fruit fly *Bactrocera zonata* (Saunders), and Mediterranean fruit fly *Ceratitidis capitata* (Wiedemann) are major insect pests infesting apricot in Egypt. The last two insect pests cause significant economic damage on a number of crops in Egypt, including apricots.

A number of aphids (e.g., the mealy plum aphid *Hyalopterus pruni*), mealybugs (e.g., the mango giant mealybug *Drosicha mangiferae*), and scale insects (e.g., the plum scale *Parthenolecanium corni*) are known to be minor pests infesting apricots (Hill, 2008).

Another important fruit crop that this research is focusing on is mango, *Mangifera indica*. The center of origin of mangoes is the Indo-Burma region, and the tree grows wild in the forests of Northeast India. Mango is now widely grown throughout the tropical regions for fruit. It is also grown in the subtropical regions as an ornamental or shade tree. The main production areas are India, US State of Florida, Egypt, Brazilian State of Rio Grande do Norte, West Indies, the Philippines, and the eastern coast of Africa. The tree is grown from sea-level to 1500 meters, but grows best below 1000 meters in climates with strongly marked seasons. Dry weather is required for flowering. The tree is susceptible to frost, and the preferred temperature is 25–30°C.

According to FAOSTAT, the world production of mangoes, mangosteens, and guavas was estimated at 38,899,593.02 tons in 2011. In Egypt, the production

was estimated at 598,084 tons in the same year, which constitutes about 1.5 percent of world production (FAOSTAT 2011 estimates). Egypt is the world's 14th largest producer of mangoes and the top producing country in the Mediterranean Basin.

In addition to *B. zonata* and *C. capitata*, which are major pests on mango in Egypt, other major pests include the coconut scale *Aspidiotus destructor* (Sign) and the pink hibiscus mealybug *Maconellicoccus hirsutus* (Green). Minor pests of mango in Egypt include, among others, the long-tailed mealybug *Pseudococcus longispinus* (Targioni-Tozzetti) and the mango soft scale *Kilifia acuminata* (Signoret).

Fruit flies are insect pests of great economic importance. There are approximately 4,000 fruit fly species, out of which around 1,200 are members of Family Tephritidae. About 40 percent of the tephritid fruit flies are polyphagous, and the remaining 60 percent attack flowers, stems, leaves, and roots of the host plant. Most of the described fruit flies belong to 5 tephritid genera: *Anastrepha*, *Bactrocera*, *Ceratitis*, *Dacus*, and *Rhagoletis*. Genus *Bactrocera* is the largest and contains about 500 described species—(Frey *et al.*, 2013; El-Heneidy, 2012).

This research is focusing on two tephritid fruit flies of major economic importance on both apricot and mango in Egypt: the peach fruit fly *Bactrocera zonata* (PFF) and the Mediterranean fruit fly *Ceratitis capitata* (Medfly).

In 1924, in his work *A Monograph of Egyptian Diptera. (Part II. Fam. Trypanidae)*, H.C. Efflatoun reported that *B. zonata* was first detected in Egypt in 1914 in Port Said, but the quarantine interception from an Indian shipment was not confirmed by further records.

According to De Meyer *et al.* (2007), the first record of *B. zonata* as an established insect pest in Egypt was in Al-Qalyubia Governorate to the east of Cairo in 1993 on samples of guava, *Psidium guajava*, and later in the same year in Al-Fayoum Governorate to the southwest of Cairo. The pest was later found to be present in most of Egypt's governorates.

As the PFF is widely spread in Egypt, it has restricted the presence of the Medfly in horticultural areas (Hashem *et al.*, 2001). It also turns out that a mixed infestation by both fruit flies would actually mostly produce PFFs regardless of which insect infested the fruit first (Mohamed, 2004). This note on mixed infestation by the two fruit flies in Egypt is of particular importance for the interpretation of the results of this study.

Control strategies for *B. zonata* in Egypt are mainly based on the use of conventional chemical pesticides. Tree trunks are either partially sprayed or baited. Killing bags are used in semi-isolated orchards and in areas with

moderate population densities. A number of cultural control methods (e.g., pruning, weeding, and collection of fallen fruits) are relatively considered by farmers for population reduction. Pheromone-mediated control measures are not a common practice (El-Heneidy, 2012).

In a related effort, a PFF eradication program in cooperation with the International Atomic Energy Agency (IAEA) and the FAO is under way in Egypt. The program utilizes the Male Annihilation Technique (MAT) as a method for eradication, and was made available to Egypt in 2000 by an IAEA-funded FAO technical cooperation project (Aleryan, 2006). It might be of relevance to mention that MAT has been found to be more effective than baiting techniques against the PFF in Pakistan (Ali *et al.*, 2010).

Fruit bagging to prevent female oviposition and quarantine measures to prevent the importation of infested fruits are recommended control measures suggested by the procedure for official control followed by the European Plant Protection Organization (EPPO, 2010).

As for the Medfly, control strategies in Egypt are mainly based on sprays of conventional synthetic pesticides (Hashem and El-Halawany, 1996). Broad-spectrum insecticides are generally used. However, it has also been observed during the field visits of this study that an extensive number of homemade traps containing a product called Buminal are used for control. Buminal is a mixture of protein hydrolysate and a generic salt, and the product is used for mass trapping purposes in traps made of plastic bottles.

Fruit stripping is a cultural control measure in which fruits are stripped from host trees, placed in plastic bags, and then buried. Another technique that proved to be successful, though it is not applied in Egypt yet, is the Sterile Fruit Fly Release, in which the orchard is flooded with sterile flies produced in rearing cages. When the sterile flies mate with the fertile population, no offspring are produced. Gradually, the wild flies can find only sterile flies to mate with, and eventually the wild population is eradicated from the agro-ecosystem (Thomas *et al.*, 2001).

In Egypt, the most commonly used control measure against the Medfly and the peach fruit fly in apricot and mango orchards is chemical sprays. Although some unconventional control measures can be found in the large areas owned by some fruit processing firms, other areas of the fragmented agricultural lands owned by "small" farmers have almost exclusively chemical sprays as a control measure. However, as mentioned earlier, it was noted during the field trials conducted for this study that some small farm owners tend to use a primitive type of traps (empty coke plastic bottles with small amounts of a local attractant) as a cheap complementary control measure besides spraying.

Egyptian exporters of agricultural crops, however, often find it difficult to strike a balance between efficient pest management requirements and export market requirements, particularly when it comes to the European export market. This is due to the restrictions imposed by importing countries on the maximum residue limits (MRLs) of pesticides.

This explains the need for applying unconventional controls against agricultural pests, with a total or near total annulment of pesticide application. Nowadays, such a need has utmost priority due to the growing concerns for public health.

Pesticide residues on harvested agricultural goods increasingly put the health of consumers and farmworkers at risk. Between October 1996 and May 1997, a market basket survey for pesticides was conducted in a Caribbean country, and the results showed that 10 percent of the surveyed produce exceeded the internationally acceptable MRLs for the respective pesticides (El-Saeid, 2003).

Also, according to estimates of the World Health Organization and United Nations Environmental Program, each year there are 1 to 5 million cases of pesticide poisoning among agricultural workers, with about 20,000 fatalities mostly reported from developing countries (El-Wakeil *et al.*, 2013).

In a recent experiment conducted in Egypt, 132 samples of fruits, vegetables, herbs, and spices collected from local markets were analyzed for pesticide residues, and contamination with pesticide residues reached 54.55 percent of the samples (72 samples), with one sample violating Codex MRLs. It is worth noting here that 6.06 percent of the contaminated samples had 2 pesticide residues, and 5.3 percent had more than 2 residues. Furthermore, 2 caraway and 1 fennel samples contained 4 pesticide residues; 1 marjoram sample contained 5 pesticide residues; and 1 mint sample contained 6 pesticide residues! Six of the pesticides detected as residues in the analyzed food items were considered to be carcinogenic (Farag *et al.*, 2011).

On the other hand, insecticide resistance (a sort of genetic changes caused by human activity) is another setback of insecticide-based controls. Excessive use of insecticides in pest control activities over the years has contributed to genetically induced resistance in many insect species, a matter which renders pest control efforts less effective.

Insecticides have been extensively used for the control of tephritid flies, which attack a large variety of fruits representing highly-priced commodities in many countries. Due to the nature of fruit flies as highly mobile insect pests with tendency for wide spatial dispersal, their insecticide resistance has been considered to be less evolving than the insecticide resistance of other insects. However, recent studies indicate that selection pressure has already reached

the point where insecticide resistance is obviously detectable in fruit flies in the field, which renders control efforts problematic (Vontas *et al.*, 2011).

Such evolving fruit fly insecticide resistance has become a problem in field situations, and a relevant example is the case of Medfly's resistance to malathion in Spain in such a way that resistance levels have overcome the insecticide concentration used in field treatments (Magaña *et al.*, 2007).

Another relevant example is a study conducted at the Faisalabad-based University of Agriculture in Pakistan on PFF resistance to a number of insecticides, where the study results showed that the fly was resistant (3 to 19 fold) to trichlorfon, malathion, lambda-cyhalothrin, and bifenthrin (Ahmad *et al.*, 2010).

From the above, it becomes obvious that there is an urgent need for an alternative approach to the strategy followed for the control of the two fruit flies. One possible solution is the use of the Attract & Kill (A&K) technique and a bait-based Male Annihilation Technique (MAT) as sustainable, eco-friendly, and effective control measures.

Therefore, the objectives of this study are laid out as follows:

- Monitoring and studying the population densities of the PFF and Medfly in apricot and mango orchards in Egypt.
- Evaluating the impact of using the A&K technique Ceranock against both fruit flies on apricot and mango orchards in Egypt.
- Evaluating the impact of using the bait-based MAT Zonatrac against the PFF on apricot and mango orchards in Egypt.

Chapter 2

Literature Review

2.1. General Information and Statistics

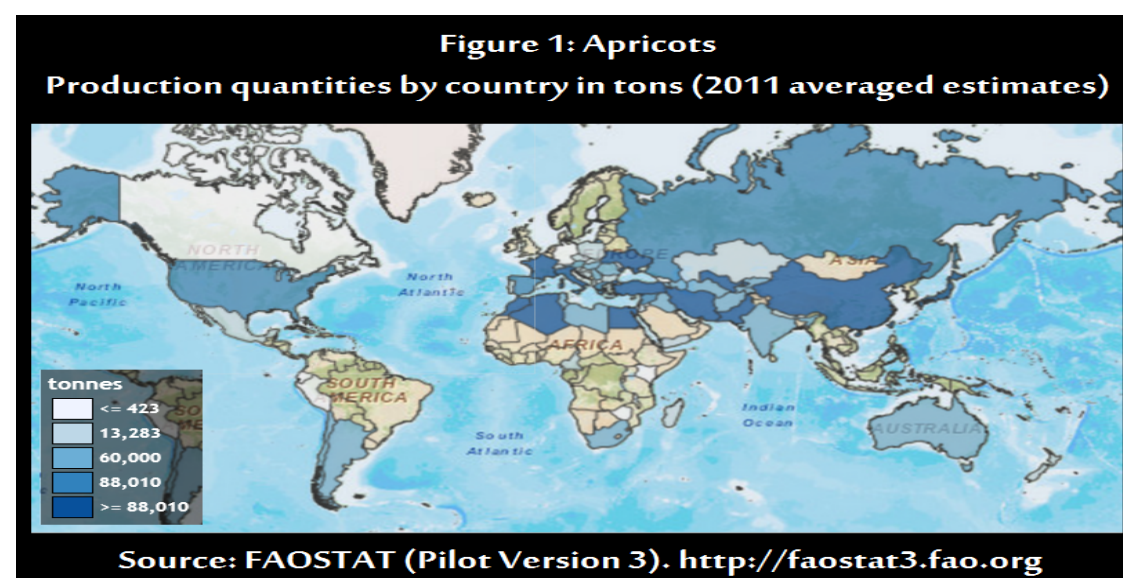
2.1.1. Apricot

Apricot, *Prunus armeniaca* (L.), is a stone fruit that belongs to family Rosaceae. Cultivated throughout the world's temperate regions, apricots are eaten fresh or cooked. While it is originally native to China, apricot is now cultivated in all countries of Central and Southeast Asia. It is also cultivated in parts of South Europe and North Africa (Fig. 1).

Turkey is the leading country in apricot production. Other important producers include Iran, Uzbekistan, Italy, Algeria, the United States, and France (see Table 1 and Fig. 1).

Table 1. Principal countries producing apricots in 2011. All volumes are in metric tons. Source: Statistical Division of the Food and Agriculture Organization of the United Nations (FAOSTAT, 2013)

Country	Production
Turkey	676,138
Iran	452,988
Uzbekistan	356,000
Italy	263,132
Algeria	205,000



Although apricot trees are susceptible to late spring frosts, they perform best in climates with dry spring weather (Fig. 1). The trees are best planted at about 10 to 20 foot spacing. In Egypt, apricots, which are mostly self-fruitful trees, ripen between mid-June and late June within 100 to 120 days from full bloom (Pittenger, 2002).

Good sanitation practices are normally necessary to control the pests of apricots: all dead or diseased wood should be cut; dried apricots should be removed from the trees; and leaves and fallen debris should be cleared away from the orchard. Before and after use, pruning tools should be disinfected with a 10-percent solution of household bleach, and the areas between trees should also be disinfected with a similar solution. In spring, a horticultural oil should be sprayed on apricot trees at the first sign of green growth for the control of scale insects and a reduction of overwintering mites and aphid eggs (University of Illinois Extension, 2013).

2.1.2. *Mango*

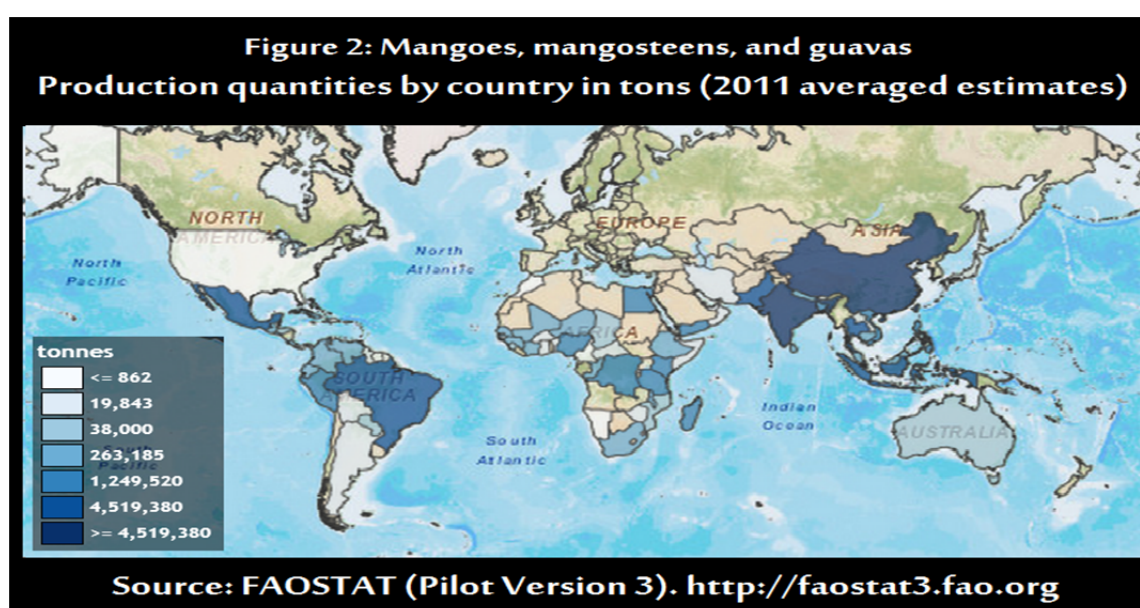
A tropical fruit belonging to the Anacardiaceae family, mango is native to eastern India and Burma. While several hundred varieties of mango exist, only a few are commercialized. Apart from bananas, the mango is the most consumed tropical fruit worldwide. More than 90 countries grow it (UNCTAD, 2012)—see Fig. 2.

Global production of mangoes has doubled in 30 years to around 35 million metric tons in 2009. Asia, which is the origin of mango, is the largest producer, with 77 percent of world production. Asia is followed by the Americas and Africa with 13 and 10 percent of world production, respectively. India, where the mango is regarded as the “king of fruits,” is the main producer worldwide, with 13 to 17 metric tons annually. China ranks second worldwide with more than 4 metric tons, followed by Thailand (2.5 metric tons) and Pakistan (1.7 metric tons). In the Americas, Mexico (1.5 metric tons) and Brazil (1.2 metric tons) rank 5th and 7th, respectively. The main African mango-producing country is Nigeria followed by Egypt, whose production is estimated at 598,084 metric tons according to FAOSTAT 2011 estimates (UNCTAD, 2012).

Mango is basically a fruit that is consumed locally. Although international trade in mangoes is constantly increasing, it still represents only 3 percent of the volumes produced. This is due to the fact that mangoes, which are delicate and easily perishable fruits, are difficult to sell (UNCTAD, 2012). Another reason is obviously the infestation of mango by fruit flies, a matter which is becoming a major problem facing mango producers, particularly in countries where the PFF is present besides the Medfly.

Table 2. Principal producers of mangoes, mangosteens, and guavas in 2011. All volumes are in metric tons. Source: Statistical Division of the Food and Agriculture Organization of the United Nations (FAOSTAT, 2013)

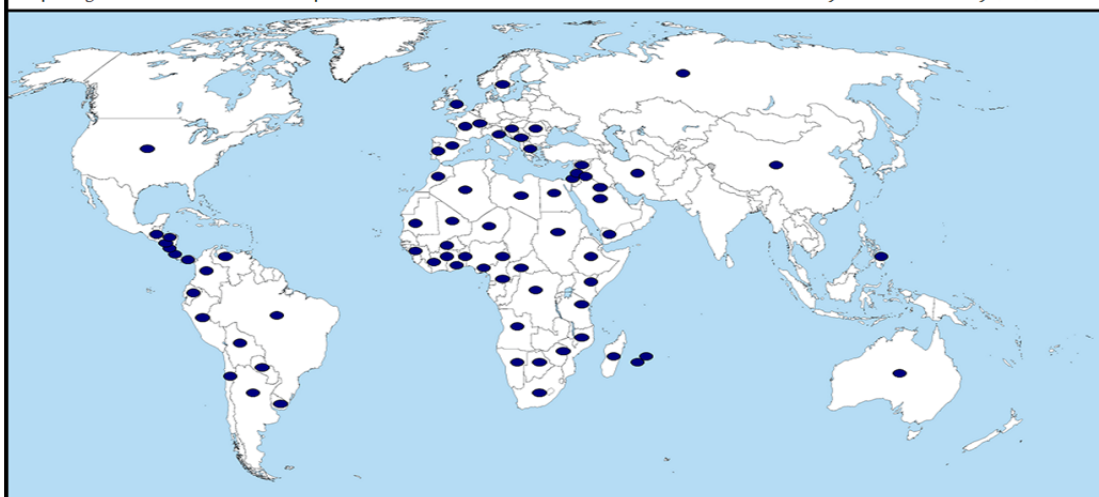
Country	Production
India	15,188,000
China	4,519,380
Thailand	3,277,250
Indonesia	2,131,140
Pakistan	1,888,450



2.1.3. Mediterranean Fruit Fly (Medfly)

A major insect pest on apricot and mango in Egypt is the Medfly. The Medfly, which originated in sub-Saharan Africa, is considered the most important fruit fly species worldwide. This is due to its worldwide distribution (a metropolitan pest), its wide range of hosts (400 hosts), and its high tolerance to cool, subtropical, and tropical climates (USDA, 2013)—see Fig. 3. Other reasons behind the massive spread worldwide include the fly's rapid dispersion mediated by the expansion of world trade, the cultivation of host plants in areas close to human habitats, and the smuggling of prohibited fruits and vegetables in violation of quarantine regulations (Bergsten *et al.*, 1999).

Figure 3: Worldwide distribution of Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann)
Map designed based on distribution maps and information from CAB International and the Global Biodiversity Information Facility (GBIF)



The fly is recorded in 132 countries and groups of islands in different areas in Africa, Asia, Central America, Europe, North America, Oceania, and South America (Commonwealth Institute of Entomology, 1984).

While the fly has a wide range of hosts including fruit and vegetable hosts, the adults can actually feed on all sorts of protein sources, including animal excreta, in order to develop eggs (Sela *et al.*, 2005).

In the Mediterranean Basin, the Medfly was detected for the first time in 1829 in the Atlantic islands Azores, Madeira, and Cape Verde. It was later reported in Spain (1842), Algeria (1859), Italy (1863), Tunisia (1885), and France (1900) (Mediouni-Ben Jemâa and Boushih, 2010). In 1904, it was reported as a pest in Egypt (Headrick and Goeden, 1996).

Fruit loss due to damage caused by the Medfly is estimated at US\$365 million in Mediterranean countries (Lysandrou, 2009). Also, in the Mediterranean Basin, it was noted that the fly causes damage particularly to citrus and peach (Cayol *et al.*, 1994). In Turkey, yield losses due to Medfly infestation are estimated to levels up to 80 percent, if no proper control measures are applied (Elekçioğlu, 2013). In Egypt, fruit damage due to Medfly infestation can be severe in some hosts more than others. A study in a reclaimed desert area in Egypt showed that Medfly infestation can reach levels up to 74 percent on apricot as opposed to only 5.7 percent on Valencia orange (Saleh and El-Hamalawii, 2004).

Another form of damage caused by the Medfly is the fact that it can transmit fungi causing fruit rot (Cayol *et al.*, 1994). A more recent study showed that the fly can even be a potential vector of human pathogens transmitted to fruits. In a lab experiment, Medflies exposed to fecal material enriched with green-fluorescent-protein-tagged *Escherichia coli* were contaminated with *E.*

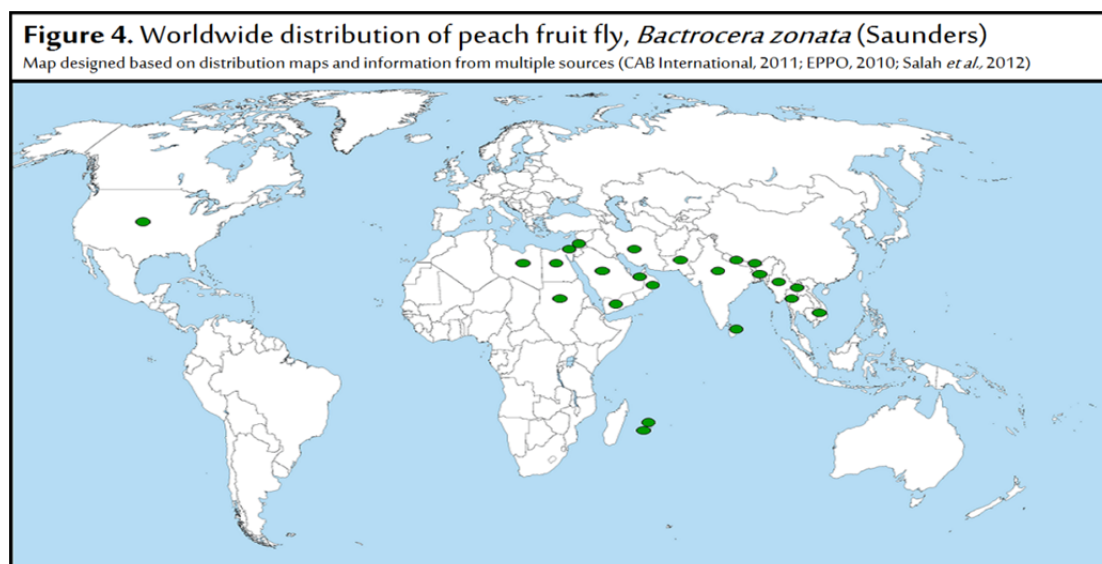
coli and were capable of transmitting the bacteria to intact apples placed in a cage. Wild flies were also found to carry coliforms (Sela *et al.*, 2005).

2.1.4. Peach Fruit Fly (PFF)

Another serious insect pest of apricot and mango in Egypt is the PFF. It causes severe damage also to peach and guava. Many other fruits and vegetables are infested by this fly. In certain areas of North India and Pakistan (the region where the pest originally comes from), the PFF has been more notorious than the Oriental fruit fly *Bactrocera dorsalis* (CAB International, 2011).

A polyphagous insect pest, the PFF has more than 50 hosts (EPPO, 2005). While its origin is South and Southeast Asia, it is now found in more than 20 countries, mostly in Asia. In the Mediterranean Basin, it is found only in Egypt and quite recently in Libya. Only very recently in July 2011, not very far from the Mediterranean Basin, the pest was found to be present in Sudan in three locations in Wad Medan of Al-Gezira State (Salah *et al.*, 2012).

Other Mediterranean/Mid-Eastern countries where the fly is present include Palestine, Saudi Arabia, Yemen, UAE, Oman, Iran (CAB International, 2011), and Lebanon (EPPO, 2010). In Saudi Arabia, the fly was introduced in 1982, and is known to be present as an invasive species in Jazan, Najran, and the southeast region of the kingdom (CAB International, 2011)—see Fig. 4.



Because of its wide range of hosts, the PFF can easily adapt and spread after being introduced into a new territory. Its establishment in a newly invaded area is also much helped by its high reproductive potential (up to 564 eggs in a lifetime), high biotic potential (several generations annually), rapid dispersal, and strong flying ability (CAB International, 2011).

The economic impact of the PFF primarily results from the loss of export markets due to quarantine measures imposed by the importing countries. In countries where the pest is present, costly quarantine restrictions and eradication measures are required by local authorities. Furthermore, the establishment of the PFF in an area can seriously impact the environment due to the initiation of chemical and biological control programs (CAB International, 2012).

Crop loss due to PFF infestation also adds to the severity of economic impact, due to the high percentage of fruit damage in infested areas. A striking example is the case in Egypt where crop loss due to the PFF is estimated at 190 million Euros a year, almost 60 percent of the annual costs of damage by the same pest in the entire Near East (320 million Euros)—(EPPO, 2005).

2.2. Use of Pheromones and Parapheromones in Pest Management

There are many chemical and visual lures that can attract insects. They are used to monitor or directly reduce insect populations either by mass trapping or through the A&K technique. Such attractants are used in ways that do not pose a threat to animals or humans as in the case of pesticides, which leave residues on foods and feeds. They are thus used in an environmentally sound manner in integrated pest management (IPM) programs (Weinzierl *et al.*, 1995).

Pheromones are semiochemicals produced and received by individuals of the same species. They influence a range of behaviors and biological processes. However, IPM programs often use compounds that attract a mate (sex pheromones) or call other individuals to a suitable food or nesting site (aggregation pheromones). Other pheromones are used to regulate the caste or reproductive development in social insects (e.g., honey bees and termites), to signal alarm (honey bees, ants, and aphids), to mark trails (ants), and to serve other functions (Weinzierl *et al.*, 1995).

Pheromone traps can be so effective for catching certain insect pests. That is why the use of a sufficient number of traps throughout a pest's environment can substantially reduce the pest's local population and limit the damage it causes. The process of placing such traps with the aim of reducing an insect pest's population is termed "Mass Trapping" or "Attract & Kill" (Weinzierl *et al.*, 1995).

The practice of combining insect attractants with insecticides has been used in IPM programs for many years. For example, poisoned bran baits were used for the control of grasshoppers in the early 1900s. The grasshoppers attracted by the treated bran were killed by an insecticide that could not be applied safely, economically, or effectively in any other way.

The process of using a high density of bait stations consisting of an insecticide combined with a lure attracting only male individuals of an insect species is termed a “bait-based Male Annihilation Technique” (MAT). The process aims at reducing the male population of an insect pest to such a low level that mating does not occur.

There are several examples of the successful use of methyl eugenol (ME) in that technique. In the 1960s, the oriental fruit fly *Bactrocera dorsalis* was eradicated using such a technique from Guam and the Commonwealth of the Northern Mariana Islands. Outstanding successes using this technique have also been achieved for the eradication of the same fruit fly from California and from the Amami Islands of Japan (Secretariat of the Pacific Community, 2002).

2.3. Mediterranean Fruit Fly, *Ceratitis capitata* (Wiedemann)

2.3.1. Taxonomic Hierarchy

Kingdom: Animalia

Phylum: Arthropoda

Subphylum: Hexapoda

Class: Insecta

Subclass: Pterygota

Infraclass: Neoptera

Order: Diptera

Suborder: Brachycera

Infraorder: Muscomorpha

Family: Tephritidae (Newman, 1834)

Genus: *Ceratitis* (Macleay, 1829)

Subgenus: *Ceratitis* (*Ceratitis*) Macleay, 1829

Species: *Ceratitis capitata* (Wiedemann, 1824)

Integrated Taxonomic Information System (ITIS, 2013)

2.3.2. Morphology

Egg. Smooth, shiny white, very slender, curved, 1 mm long. The micropylar region is distinctly tubercular (Fig. 5).

Larva. White; cylindrical shape typical of fruit fly larvae; elongate; anterior end narrowed, somewhat recurved ventrally; anterior mouth hooks; flattened caudal end. The last instar is usually 7 to 9 mm in length, with 8 ventral fusiform areas. The anterior buccal carinae are usually 9 to 10 in number. The anterior spiracles are usually nearly straight on dorsal edge of tubule row (often more straight than illustrated in Fig. 5). There are usually 9 to 10 tubules, although there may be 7 to 11.

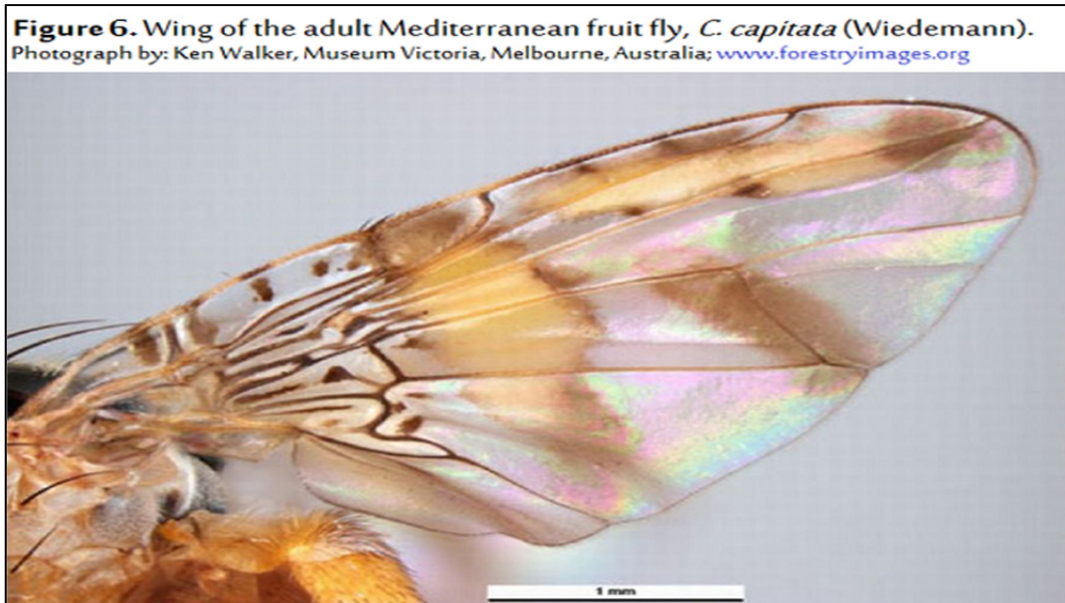
Pupa. Cylindrical; 4 to 4.3 mm long; dark reddish brown; resembles a swollen grain of wheat (Fig. 5).

Adult. Slightly smaller than a house fly; picture wings typical of fruit flies. The adult fly is 3.5 to 5 mm in length. The color is yellowish with brown tinge, especially on abdomen, legs, and some markings on wings. The lower corners of the face have white setae. Eyes are reddish purple (fluoresce green, turning blackish within 24 hours after death). Ocellar bristles are present (Fig. 5).

The male has a pair of bristles with enlarged spatulate tips next to the inner margins of the eyes. The thorax is creamy white to yellow with a characteristic pattern of black blotches. Light areas have very fine white bristles. Humeral bristles are present. Dorsocentral bristles are anterior of the halfway point between supraalar and acrostichal bristles. The scutellum is inflated and shiny black. The abdomen is oval with fine black bristles scattered on dorsal surface and two narrow transverse light bands on basal half (Thomas *et al.*, 2001).



Wing. Wings are usually held in a drooping position on live flies. They are broad and hyaline with black, brown, and brownish yellow markings. There is a wide brownish yellow band across the middle of the wing. The apex of the wing's anal cell is elongate. There are dark streaks and spots in the middle of wing cells in and anterior to anal cell (Fig. 6).



The males are easily separated from all other members of this family by the black pointed expansion at the apex of the anterior pair of orbital setae. The females can be separated from most other species by the characteristic yellow wing pattern and the apical half of the scutellum being entirely black. The female's extended ovipositor is 1.2 mm long.

2.3.3. Biology

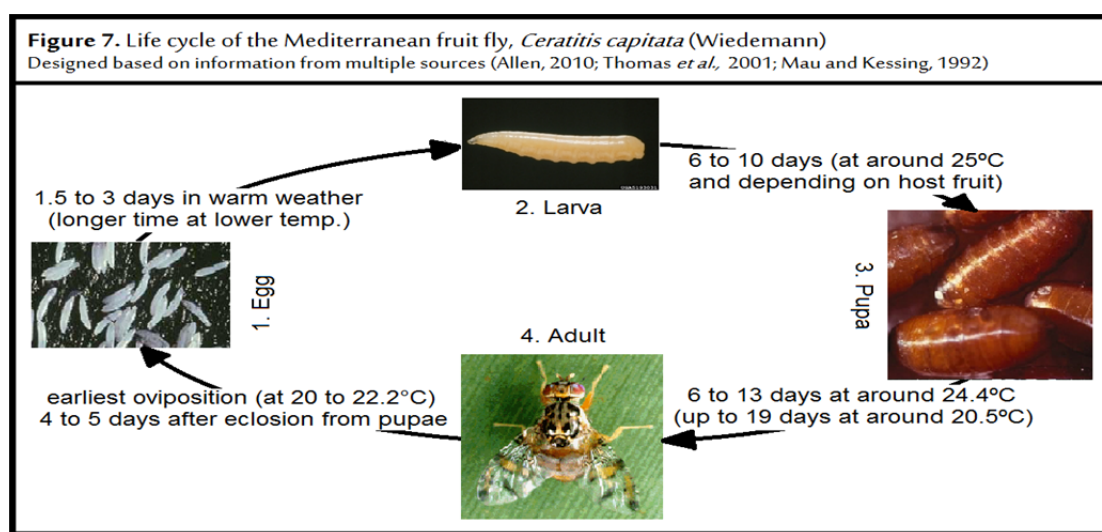
Medflies undergo a complete metamorphosis: the flies begin their life as larvae and then transform into completely different-looking adults. Mated females lay their eggs in host fruits approximately 1 mm beneath the pericarp. A female lays only 2 to 10 eggs in the one fruit. However, more than one female can lay eggs in the same location, so that the slim eggs may be clustered together in a single spot of 75 eggs or more (Allen, 2010).

Eggs hatch after 1.5 to 3 days (longer if the temperature is lower). The larvae then carve tunnels, eating their way through the host fruit. Larval life may last only 6 to 10 days (when temperature is around 25°C). Besides temperature, the type of host fruit affects the length of the larval stage. In citrus fruits, 14 to 26 days may be required for the larvae to reach pupation. Development in a green peach is completed within 10 to 15 days (Thomas *et al.*, 2001).

There are three larval stages or instars. In the first stage, larvae are slender, cream colored, translucent, and about 0.1 cm long. In the second stage, larvae are partly transparent, revealing the fruit in the gut. By the third stage, larvae are opaque white and 0.6 to 0.8 cm long. Medfly larvae can be distinguished from other fruit fly larvae by their thoracic spiracles, with 7 to 11 small protruding tubules (Allen, 2010).

Most larvae begin to pupate at sunrise, an inch or two into the soil. The pupal stage lasts from 6 to 13 days at around 24.4°C. This range significantly increases (possibly to about 19 days) when the temperature drops to around 20.5°C. The pupal stage is resistant to temperature extremes and desiccation, so it may last much longer if conditions are not right for emergence.

It is typical for the new adult Medflies to surface on warm mornings. At this early adult stage, they are capable of flying short distances, and may disperse further distances via the wind. (Mau and Kessing, 2007; Thomas *et al.*, 2001)—see Fig. 7.



2.3.4. Damage

C. capitata is a serious pest to many crops. Damage results from:

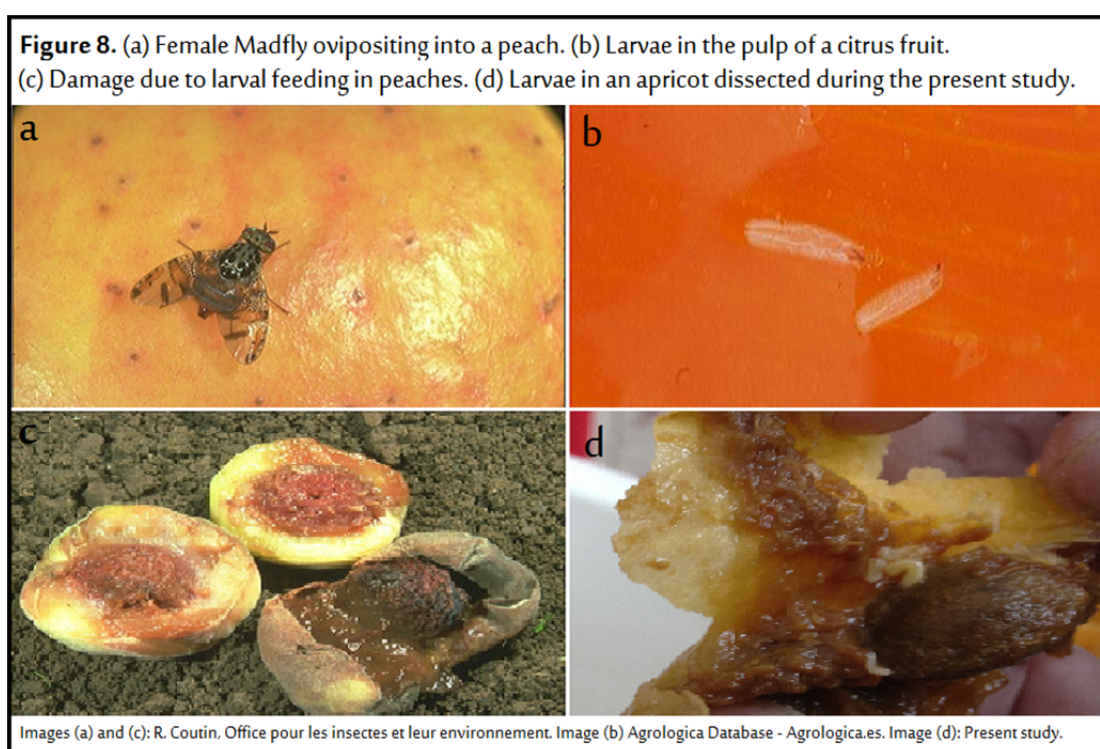
1. Oviposition in fruits and soft tissues of vegetative parts of certain plants;
2. Larval feeding;
3. Plant tissue decomposition by secondary microorganisms.

Larval feeding inside fruits is the most severe type of damage. Attacked ripened fruits may develop a water soaked appearance. Young fruits, however, become distorted and usually drop on the ground. The tunnels resulting from larval feeding serve as entry points for bacteria and fungi, which cause fruit rotting (Fig. 8). Medfly larvae also attack the host plant's young seedlings, succulent taproots, stems, and buds.

Trapping for population detection and population exclusion (using foliage baits), as well as chemical sprays and releases of sterile male Medflies to reduce populations, all require a wide range of resources and can have significant economic implications. Medflies are serious quarantine pests that

also affect world trade. The presence of Medflies often calls for quarantine treatments or disinfestation measures for areas of host crops to be certified as fly-free in certification programs. The costs of such phytosanitary regulatory measures can be significant (Global Invasive Species Database, 2010).

As *C. capitata* is polyphagous, it takes advantage of the various hosts in its surrounding environment and uses them as stepping stones to move on from one fruit tree species to another as fruits mature throughout the season. This eventually gives the flies the ability to destroy an area's entire production of many fruits. Also, such ability to infest multiple fruit species provides Medflies with refuges from control measures, as different fruit species serve as a source of re-infestation to surrounding plots (ibid).



2.3.5. Integrated Management of *C. capitata*

Mechanical Control. One of the most effective mechanical control methods is bagging the fruits to exclude egg laying. Trapping is an alternative method that is yet to be proved completely effective.

Cultural Control. The principal cultural control method used for controlling this pest is field sanitation directed toward the destruction of all unmarketable and infested fruits. Infested fruits should be buried 3 feet under soil surface with an addition of sufficient lime to kill larvae. Weekly harvesting keeps the quantity of ripe fruits on the trees to a minimum, thus reducing food sources

from which large populations may develop. Other practices that reduce the amount of in-field breeding of Medflies should be used.

Biological Control. According to Mau and Kessing, between 1947 and 1952, 32 entomopathogenic species and varieties of natural enemies to fruit flies were introduced to Hawaii. These parasites lay their eggs in the eggs or maggots of fruit flies and emerge in the pupal stage. Only three—*Opius longicaudatus* var. *malaiaensis* (Fullaway), *O. vandenboschi* (Fullaway), and *O. oophilus* (Fullaway)— have become abundantly established. These parasites are primarily effective on the oriental and Mediterranean fruit flies in cultivated crops.

A number of other parasites have also been introduced into Hawaii specifically for Medfly control. The most important were the braconid wasps *Opius humilis* and *Diachasma tryoni*. Later, parasites of the Oriental fruit fly were found to be destroying the Medfly. They are *Biosteres oophilus*, *B. vandenboschi*, and *B. longicaudatus* listed in order of their effectiveness.

Chemical Control. Chemical sprays have not been completely effective in protecting fruit crops from Medflies. Egg laying requires only a few minutes and chemical residues do not kill adults within this time frame.

The use of proteinaceous liquid attractants in insecticide sprays is a recommended method for controlling adult Medfly populations in the vicinity of crops. Insecticide-bait sprays are applied to broad leaf plants that serve as a refuge for adult Medflies. Baits serve to encourage the adults (especially females) to feed on the spray residue, and can provide good rates of killing. To be effective, insecticide-bait sprays must be used in combination with good sanitation practices (Mau and Kessing, 2007).

2.4. Peach Fruit Fly, *Bactrocera zonata* (Saunders)

2.4.1. Taxonomic Hierarchy

Kingdom: Animalia

Phylum: Arthropoda

Subphylum: Hexapoda

Class: Insecta

Subclass: Pterygota

Infraclass: Neoptera

Order: Diptera

Suborder: Brachycera

Infraorder: Muscomorpha

Family: Tephritidae (Newman, 1834)

Genus: *Bactrocera* (Macquart, 1835)

Subgenus: *Bactrocera* (*Bactrocera*) Macquart, 1835

Species: *Bactrocera zonata* (Saunders, 1842)

Integrated Taxonomic Information System (ITIS, 2013)

2.4.2. Morphology

Color. Face with a spot in each antennal furrow; scutum with lateral yellow or orange vittae; scutellum entirely pale colored, except sometimes for a narrow black line across the base; costal margin of wing without a colored band along whole length of cell r1; cell sc usually yellow, and apex of vein R4 + 5 often with a brown spot; crossveins R-M and Dm-Cu not covered by any markings (Fig. 9).

Head. With reduced chaetotaxy, lacking ocellar and post-ocellar setae; first flagellomere at least three times as long as broad (Fig. 9).

Thorax. With reduced chaetotaxy, lacking dorsocentral and katepisternal setae. Post-pronotal lobes without any setae (sometimes with some small setulae or hairs); scutum with anterior supraalar setae and prescutellar acrostichal setae; scutellum not bilobed, with only two marginal setae (the apical pair)—see Fig. 9.

Abdomen. All tergites are separate (view from side to see overlapping sclerites); tergite five with a pair of slightly depressed areas (ceromata); male with a row of setae (the pecten) on each side of tergite three (Fig. 9)—(EPPO, 2005).

Figure 9. Dorsal view of a complete adult (top left), head (top right), thorax (bottom left), and abdomen (bottom right) of the peach fruit fly, *Bactrocera zonata* (Saunders)

Source of Images: Ken Walker, Museum Victoria. MuseumVictoria.com.au



Wing. Vein sc is abruptly bent forward at nearly 90°, weakened beyond this bend, and ending at subcostal break. Vein R1 with dorsal setulae; cell bcu (= cup) is very narrow, about half depth of cell bm; bcu (= cup) extension is very long, equal to or longer than the length of vein A1 + CuA2; 4–6 mm long. Raised narrow subbasal section of cell br lacking microtrichiae (Fig. 10).

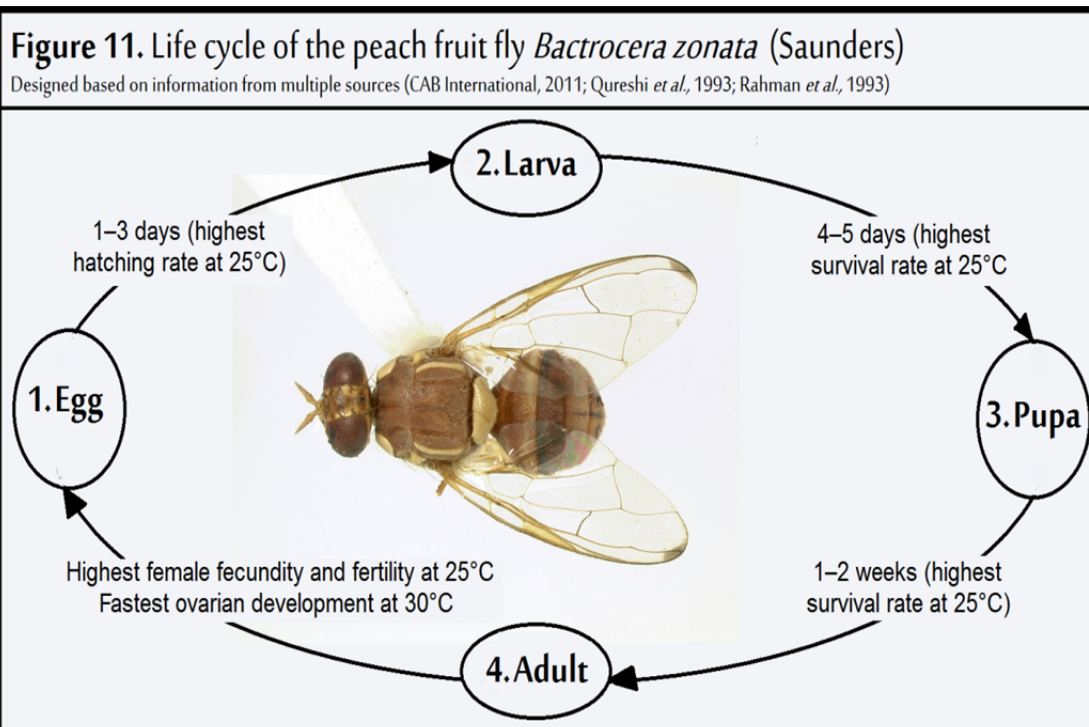


2.4.3. Biology

The flies overwinter in the pupal stage, and then adults emerge with the increase of ambient temperature and start mating. After selecting a suitable site for oviposition, a mated female inserts her ovipositor into the host fruit beneath the pericarp and deposits 3 to 9 eggs at one time.

Eggs then hatch within 1 to 3 days, and the larvae feed on the fruit tissue and grow for another 4 to 5 days inside the host fruit. The duration of various immature stages normally varies at different temperatures. No stages develop at 15°C or less, and the optimum temperature is 25 to 30°C (Qureshi *et al.*, 1993).

Full-grown larvae enter the soil under the host plant for pupation, and then adults emerge after 1 to 2 weeks (longer in cool conditions). Adults occur throughout the year. The eclosion of adults from pupae mainly occurs in the early hours of the morning (CAB International, 2011; Rahman *et al.*, 1993)—see Fig. 11.



For most *Bactrocera* spp., adults are the only life stage that can best survive low temperatures, with a normal torpor threshold of 7°C. Temperatures survived can even drop as low as 2°C in winter, which explains why the fly is now established in Egypt after it had been originally considered an exclusively tropical fruit fly. This raises questions about the fly's possible survival during cold winters (EPPO, 2005).

2.4.4. Damage

Signs of oviposition punctures usually appear on attacked fruits. Fruits with high sugar content, such as peaches, exude a sugary liquid droplet that usually solidifies adjacent to the oviposition puncture. The dry droplet appears in the form of a brown, resinous deposit (EPPO, 2005).

On hatching, larvae eat their way into the interior of the host fruit. The activity of first-instar larvae is restricted to the area below the oviposition puncture. However, second- and third-instar larvae are voracious feeders: they go deeper in the host fruit tissue and are responsible for the complete deterioration of host crops (CAB International, 2011)—Fig. 12 and Fig. 13.



2.4.5. Integrated Management of *B. zonata*

Sanitary Measures. Proper field sanitation is essential. Infested host fruits are plucked and those on the ground are collected and buried deep in the soil. After harvest, if some fruits are left unpicked on the trees they become the source of later infestation, so all fruits should be picked (Plantwise, 2013).

Physical Control. This type of control is mainly based on the wrapping or bagging of individual fruits to prevent female oviposition. It has proved to be effective (CAB International, 2011).

Chemical Control. Chemical controls based on bait sprays and on relatively less hazardous insecticides like malathion seem to be the most efficient control methods available (Roessler, 1989).

Bait-Based Male Annihilation Technique (MAT). Methyl eugenol is an effective attractant to PFF males. It is mixed with an insecticide and protein bait and used in traps. Attractant-based male annihilation can be effective in substantially reducing an insect's population area-wide if it is carried out on a large scale. It is also worth mentioning that MAT is officially considered as part of the eradication treatments recommended by the European and Mediterranean Plant Protection Organization (EPPO).

A treatment used by the EPPO procedure for official control of *B. zonata* is aerial proteinaceous bait sprays. Supplemental eradication methods include ground-applied proteinaceous bait sprays, soil treatment with diazinon, and stripping and disposal of ripe fruits within 200 meters of a confirmed larval site (EPPO, 2010).

Plant Quarantine. Prevention of the PFF from establishing in areas free from the pest is achieved through strict quarantine regulations. Imports of host fruits and vegetables from infested areas without post-harvest disinfestation measures should be prohibited. Thorough checking of travelers' baggage for infested hosts at entry ports is also essential.

Post-harvest Treatment. Many countries forbid the import of host fruits without a strict post-harvest treatment applied by the exporter in advance. Such treatments include fumigation, heat treatment (hot vapor or hot water), cold treatments, insecticidal dipping or irradiation (Armstrong and Couey, 1989; Armstrong, 1997). Many countries now do not accept both irradiation and methyl bromide fumigation. Heat treatment can reduce the shelf-life of most fruits, and therefore the most effective method of regulatory control is the restriction of imports of a given fruit to areas free from the fly (CAB International, 2011).

2.4.6. *B. zonata* in Egypt

In 1924, *B. zonata* was declared present in Egypt. The declaration was based on a detection of the fly in an intercepted consignment in Port Said in 1912. The pest was no longer mentioned for a long period of time, until an intensive tephritid fruit fly survey was initiated by the FAO in the 1980s, but the PFF was not found then. In 1998, the pest was identified for the first time on infested guavas collected in Al-Agamy and Al-Sabahia districts near Alexandria (EPPO, 2013).

In 1999, the first traps that were set up for the fly showed high capture rates in Alexandria and Cairo. In October 2000, the PFF was detected in Al-Arish in North Sinai. A monitoring scheme set up in the North Sinai Governorate involved the installation of 45,000 A&K blocks.

At present, the PFF is considered present and widespread in Egypt, and the situation can be detailed as follows. Mainland: whole Nile Delta region, Nile Valley, and Al-Kharga and Al-Dakhla oases. There are extremely high populations in Cairo (>30 flies per trap per hour in downtown Cairo). Sinai peninsula: Ras Sidr, Al-Tur, and Nuweiba in the South Sinai Governorate. Captures all along the North Sinai Governorate (130 km² of potential hosts) from Al-Qantara in the northwest to Rafah in the northeast. High populations are found in gardens in Al-Arish city of the North Sinai Governorate (EPPO, 2013).

The pest is also present on the Egypt-Palestine border south of Rafah City. No efficient control action has yet been taken. It is stressed that the pest is present even in very dry areas with few host plants, and even on isolated trees. While the fly is found in peach, apricot, and mango orchards, larger populations occur in gardens with several different fruit trees in a relatively limited area. Although eradication seems to be difficult to achieve nationwide, it appears to be achievable in the Sinai Peninsula (EPPO, 2013).

2.5. Background of the Study's Control Strategies

Over the twentieth century, different traps and attractants have been developed and applied for the purpose of surveying fruit fly populations. Historically, methyl eugenol (ME) was the first attractant to be used exclusively for male fruit flies in what is referred to now as a bait-based MAT. ME was then followed by kerosene as an attractant to Medflies. In 1956, Angelica seed oil was also utilized as an attractant to Medflies (IAEA, 2003).

Trimedlure (TML) was later found to be an effective attractant to Medfly males (Beroza *et al.*, 1961). Two years later, Beroza and Green (1963) demonstrated that cuelure is an effective attractant to *Bactrocera cucurbitae*.

Other food baits based on protein solutions, fermenting sugar solutions, fruit juices, and vinegar have been in use since 1918 as attractants for the females of a number of fruit fly species (IAEA, 2003).

Different types of traps are currently used worldwide (see examples in Fig. 14 and Fig. 15). For fruit fly surveys, such traps complement fruit fly control activities and eradication campaigns.

The first trap to be used with protein baits was the McPhail trap. It was later followed by the development of Steiner traps in 1957 and Jackson traps in 1971. While McPhail traps are often used with protein attractants, Jackson traps are used with TML and Steiner traps with ME or cuelure (IAEA, 2003).

Figure 14. The flycatcher or McPhail trap (left) and mothcatcher (right) for pheromone- and parapheromone-mediated pest management.
Images courtesy of Russell IPM Ltd.

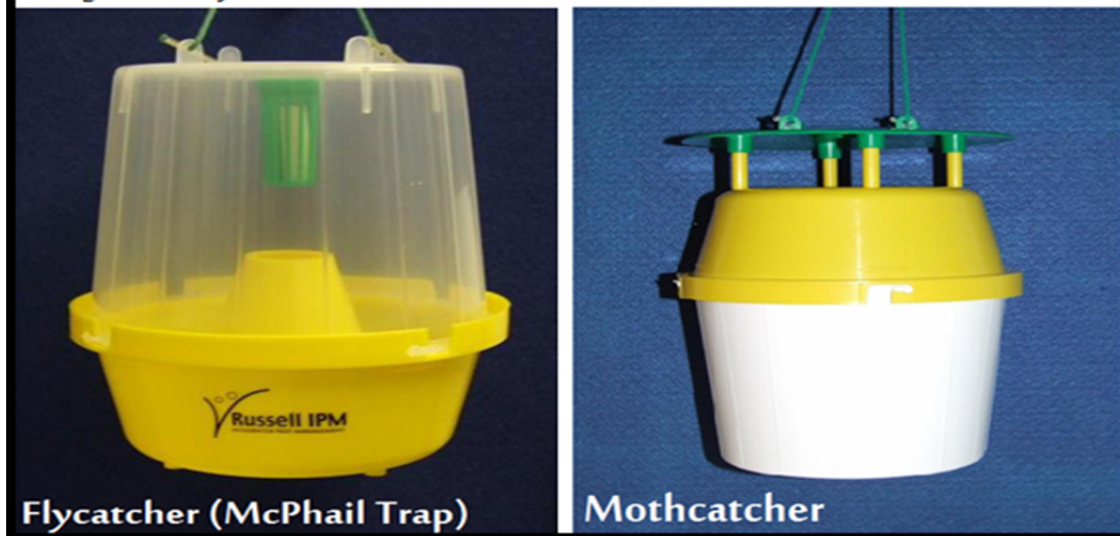
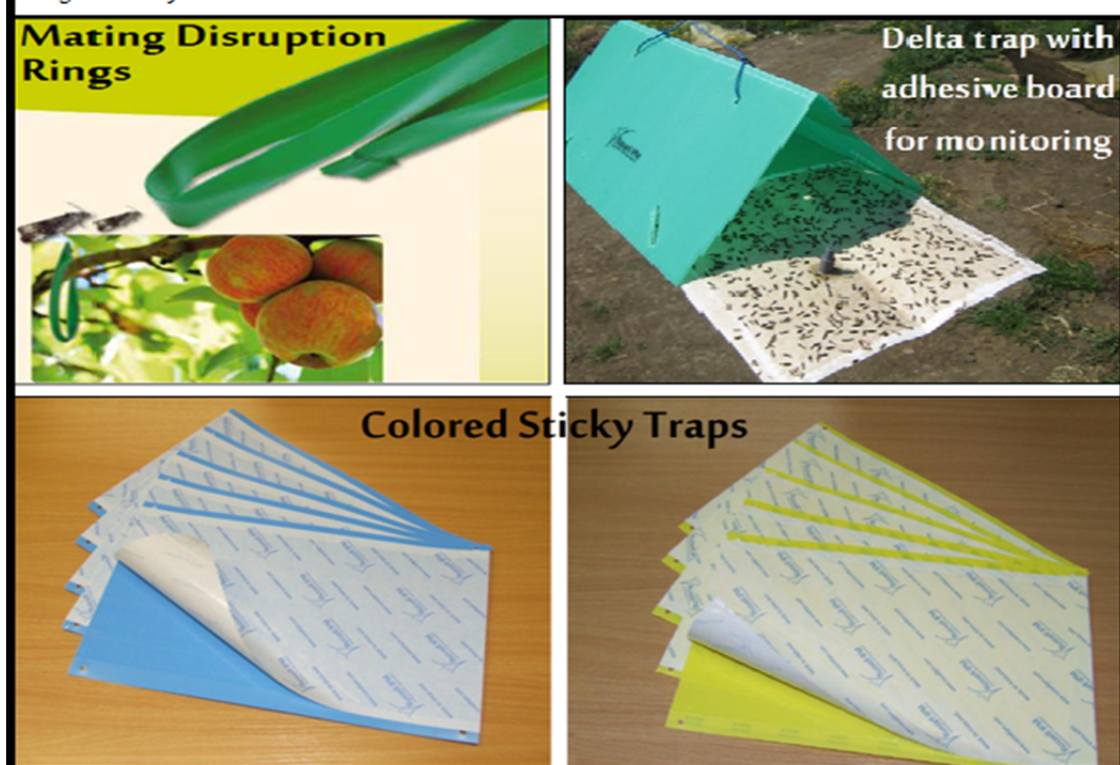


Figure 15. Mating disruption rings, delta trap (monitoring), and colored sticky traps (mass trapping) as pheromone- and parapheromone-mediated techniques for pest management.
Images courtesy of Russell IPM Ltd.



Traps used for fruit flies depend on the nature of the attractant. The most widely used traps contain parapheromone or pheromone attractants that are male specific. The parapheromone TML captures the Medfly and Natal fruit fly. The parapheromone ME captures a large number of *Bactrocera* species, including the PFF. The parapheromone cuelure also captures a large number

of *Bactrocera* species including *B. cucurbitae*. The pheromone Spiroketal (SK) captures *B. oleae*.

Parapheromones are highly volatile compounds. They can be used with panels, delta-traps, and bucket-type traps (e.g., the McPhail trap). TML, ME, and cuelure are produced in controlled release formulations that provide a longer-lasting effect for field use.

Several synthetic food-based attractants have been developed using ammonia and its derivatives. The combination of ammonium carbonate or ammonium acetate with putrescine and trimethylamine results in a highly effective female attractant for the Medfly. This synthetic food-based attractant is used in early-detection trapping networks. A similar compound was used for the monitoring scheme of this study under the commercial name Femilure. Such attractants are more specific than liquid protein baits and can detect female Medflies at a lower level when compared to the male-specific attractant TML (IAEA, 2003).

2.6. Control Strategies of the Study

2.6.1. Principles of the A&K Technique

While the technique is referred to in the literature using different terms—such as "bait sprays" and attracticides—still the name "Attract and Kill" or "Lure & Kill" seems to be the most popular. The A&K technique has been in use by both farmers and large agricultural firms for several decades. The strategy is based on pheromone- and parapheromone-mediated control and is used in pest control and eradication programs that put environment conservation as one of its goals.

Either crude baits or synthetic semiochemicals can be used as attractants used in the traps utilized by the A&K strategy. While crude baits are extensively used against crawling insects like ants and cockroaches, semiochemicals are mainly used against flying insects of the Lepidoptera, Diptera, and Coleoptera orders (Beroza and Green, 1963).

It is worth mentioning here a crucial difference between the A&K technique and the mass trapping technique. In the A&K technique, insects are attracted to the source of the attractant but they are not entrapped there as in the case of mass trapping. What happens is that insects attracted to the lure are killed by the killing agent, and thus end up falling somewhere on the orchard's floor, which significantly reduces the target insect population (El-Sayed *et al.*, 2009). This is exactly the case with the two products used as A&K techniques in this study.

The factors affecting the success of this technique in pest management can be laid out as follows:

1. The way through which insects will be in contact with the killing agent, which can be mixed with the attractant or applied near the source of the attractant. The more feasible the bait design in such a way that allows easy and direct contact between the insect and the killing agent, the more successful the pest management will be.
2. Adequate dose of the killing agent, which will reduce the chances of the insect's leaving the attractant without getting killed. The level of mortality and adverse behavior-modifying effects eventually detrimental to the insect population are determined by the choice of an adequate dose.
3. Other factors include the placement, height, design, size of the bait station. There are also complex interactions between the attractant, insecticide, and insect behavior. Such interactions should be understood if high mortality rates are to be achieved.

The attractant's ability of attracting a significant number of the target insect population and causing them to land on and contact with the toxic substance contained in the bait should be maximized. It should not be compromised by the presence of the insecticide or other formulation components such as the gel, oil, adhesive, or plastic structure of the bait station (El-Sayed *et al.*, 2009).

Careful choice of the insecticide used as a killing agent is paramount, particularly to avoid problems like repellency, which can repel insects away from the attractant. While pyrethroids such as cyfluthrin, cyhalothrin, cypermethrin, furathiocarb, and permethrin have shown to be fast-acting insecticides, some pyrethroids are known to be repellent for some insects. However, researches on the A&K approach have been able to develop formulations that came to overcome that problem of repellency (Brockerhoff and Suckling, 1999).

Performing a post-trial analysis of each A&K technique in the framework of a research and development program can shed light on the key reasons for success or failure of the technique. Several examples in the literature have shown that the use of the A&K technique as a pest control strategy had successfully reduced insect populations and fruit damage in treatment plots.

Examination of such cases has shown a number of key reasons for the success of A&K programs:

1. Low density of the target population. There is obviously a direct relationship between the target insect's population density and the number of baits to be applied in an A&K program. This actually highlights the importance of applying monitoring traps at an adequate date prior to the application of the A&K baits, for an adequate assessment of the insect population and the equivalent number of baits needed for efficient control.
2. Attractant's competitiveness compared to wild females. Research and development trials on attractants need to ensure a high competitiveness of the attractant in comparison to wild females.
3. Optimal attractant placement and deployment prior to male emergence and throughout the flight period (Ebbinghaus *et al.*, 2001).

In turn, the cases where A&K programs failed to provide a major reduction in pest population or fruit damage also refer to key reasons behind that failure (Downham *et al.*, 1995):

1. A remarkably high density of the target insect population, in which case the insect population greatly outnumbers the bait stations placed in the treatment plots. It is worth noting here that an increase in the number of bait stations in that case would commercially be an uneconomic decision.
2. Non-isolation of the target insect population. The apricot trial of the current study is actually an immediately relevant example: the treatment plots were located amid large areas of fragmented agricultural lands cultivated with different fruit and vegetable crops. In that case there is a high risk of immigration/mobility of the target insect population from areas cultivated with trees known to be hosts of the target insect. Such mobility of insect pests is normally intensified when the areas around the area of the A&K program are sprayed with repellent insecticides.
3. Use of an inadequate attractant in terms of competitiveness in comparison with wild females.
4. Insufficient density of attractant sources in comparison to the target insect density.
5. Late application of the A&K program.

2.6.2. Examples of A&K Systems

A number of A&K techniques have been developed, particularly for the Medfly. These techniques were later commercially developed, with commercial trials advertised and marketed to the public in a variety of formulations.

GF-120 Fruit Fly Bait. The GF-120 is a spinosad-based fruit fly bait. It has been developed as a primary tool for the control and eradication of tephritid fruit flies. A prepackaged concentrated bait with a low application rate, GF-120 has a reduced risk of the killing agent with regard to both mammals and non-target insects. The killing agent in GF-120 used to be the organophosphate malathion (Roessler, 1989), which is well known to have a significantly negative effect on beneficial insects (Hoelmer and Dahlsten, 1993). Later, it was demonstrated through field tests that spinosad-based baits can replace protein baits mixed with malathion. Spinosad-based baits have shown to be providing significant control of *C. capitata* and the Mexican fruit fly *Anastrepha ludens* (Loew) in Hawaii and Florida (Burns *et al.*, 2001).

Spinosad is a selective insecticide produced through the fermentation of the naturally occurring soil bacterium *Saccharopolyspora spinosa*. According to Stark *et al.* (2004), it has an "extremely favorable mammalian and environmental toxicity profile." In addition to spinosad, GF-120 contains a mixture of sugar, protein, ammonium acetate, and other ingredients. The fact that GF-120 is applied at low rates, as well as the product's low toxicity, results in large margins of safety (Adán *et al.*, 1996).

GF-120 is known to be used for the control of many fruit fly species: Apple maggot *Rhagoletis pomonella* (Walsh); Caribbean fruit fly *Anastrepha suspensa* (Loew); cherry fruit fly *Rhagoletis cingulata* (Loew); Mediterranean fruit fly *Ceratitis capitata* (Wiedemann); melon fruit fly *Bactrocera cucurbitae* (Coquillett); Mexican fruit fly *Anastrepha ludens* (Loew); olive fruit fly *Bactrocera oleae* (Rossi); oriental fruit fly *Bactrocera dorsalis* (Hendel); Queensland fruit fly *Bactrocera tryoni* (Froggatt); South American fruit fly *Anastrepha fraterculus* (Wiedemann); and walnut husk fly *Rhagoletis completa* (Cresson). It is also effective on many fruit tree crops, especially citrus, peach, pear, and olive (Revis *et al.*, 2004).

The approach of GF-120 application is based on strategically placing large droplets to be found by the flies in their normal search for food. Uniform coverage here is not critical as in the case of conventional sprays. The bait provides its best performance when it is kept concentrated, not excessively diluted. In this respect, the standard dilution recommended for the control of tephritid fruit flies in the field is 80 ppm, which is equivalent to 1 GF-120:1.5 water (Wang *et al.*, 2005).

While GF-120 is generally applied in a so-called ultra-low-volume application, it is applied in larger droplets for Medfly control (Vargas *et al.*, 2001). Obviously, larger droplets of 5 or more millimeters in diameter can help extend the product's viability in the field for longer periods of time. During the application, active spinosad is applied at a rate of only 0.19 to 0.38 grams per hectare. Such ultra low rates of active spinosad application are made possible by the fact that the flies are attracted in large numbers to the GF-120 droplets, which results in the consumption of large quantities of the fruit fly bait (Piñero *et al.*, 2011).

As for the best time of application, GF-120 should be applied as soon as monitoring traps start catching adult flies. Alternatively, the application can start 2 to 3 weeks before fruit color break.

However, it is also worth mentioning that GF-120 commercial products can have some drawbacks due to the fact that they are based on spinosad. Direct contact with fresh GF-120 was found to be moderately harmful to harmful to 78 percent (lab studies) and 86 percent (field studies) of 25 parasitoid species tested (Wang *et al.*, 2005). Among the parasitoids to which spinosad-based products can be harmful are three major parasitoids belonging to Family Braconidae of Order Hymenoptera, and used against tephritid fruit flies: *Fopius arisanus*, *Diachasmimorpha tryoni*, and *Psytalia fletcheri* (Bouagga, 2012).

Spinosad-based products can also be highly toxic to aquatic invertebrates, with a potential for runoff, which is why it is advised not to have them applied immediately after rainfall or when there is a forecast for rain within 48 hours after application (Wang *et al.*, 2005). Another disadvantage is the product's cost of application, which is deemed expensive because application is required every 7 days. GF120 is also reported to have initiated fungal infestation in citrus orchards in Spain (M.N. Hassan, pers. comm.).

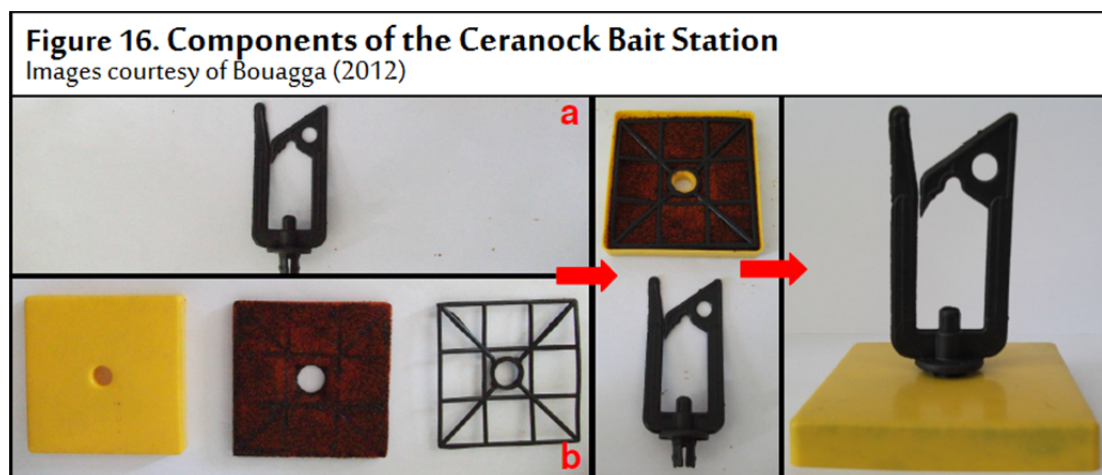
Magnet MED. Supplied by Suterra Europe Biocontrol, S.L., headquartered in Barcelona, Spain, Magnet MED is an A&K device that is basically a yellow cylindrical tube containing a mixture of a protein bait and cypermethrin at the bottom. Several small holes around the bait container allow the release of the attractants. Inside the tube, two mesoporous dispensers contain ammonium acetate, trimethylamine, and methylpyrrolidine (Navarro-Llopis *et al.*, 2013).

In trials on citrus and peach in Italy and Spain, Magnet MED has shown that it offers an innovative way as an alternative to the use of conventional insecticides for an effective control of the Medfly. Like other commercial products, it represents a fully integrated solution combining both the attractant and killing agent in one easily applicable device. Applied at a rate of 50 to 75 devices per hectare, the system has no separate traps—the attractants and

killing agent are in the same device, and no components need to be assembled (Casagrande, 2009). However, also like other products, Magnet MED has the drawback of possible damage by rain as it is made of paper. Another economic drawback is the cost of the product and its shipment fees to countries other than the product's country of origin (Bouagga 2012).

Ceranock A&K. Developed in 2011 by Russell IPM Ltd., headquartered in Flintshire, UK, the Ceranock bait station is an A&K system designed for the control of the Medfly by means of attracting and killing female flies of the population. This approach to Medfly control aims at substantially reducing the fruit fly population area-wide, with a view to reducing fruit damage and crop loss. The Ceranock A&K system works as a total or partial replacement of the chemical sprays commonly used as a means of control against the pest in many Third World countries.

The system is made up of three parts: a plastic hook used to hang the other parts on a tree branch or twig, a plastic case, and a sponge impregnated with the attractant and the killing agent. The sponge is fastened to the plastic case with a plastic net—all plastic parts are made of polyurethane & polyethylene (Fig. 16).



The attractant used in the system is a protein hydrolysate and a citrus plant extract, and the killing agent is alpha-cypermethrin. This latter is a widely used pyrethroid insecticide and is effective by contact and ingestion against target pests.

The system, being ready for use and easy to set, provides a fast solution accessible for use by all farm personnel as a control measure against the Medfly. With a lifetime of up to 4 months under normal conditions, the Ceranock A&K system can provide early control for the early fruit varieties likely to suffer more from Medfly infestation in some parts of the world. In accordance with the manufacturer's recommendation, a total of 360 Ceranock

bait stations per hectare should be applied in citrus orchards, while the number is increased to 400 stations per hectare for other fruit tree crops.

As with the case of other commercial products designed for the same purpose, the Ceranock system has a range of advantages as an IPM-compatible control measure:

- Ensuring that no pesticide is directly applied to fruit, which—besides being safe for human, plant, and animal lives—also contributes to environmental conservation efforts.
- Reducing significantly (and even replace) the practice of pesticide application as a control measure, which would in turn contribute to a reduction in pesticide resistance. This also contributes to a fruit production free from pesticide residues.

It is worth noting that the system has proved to be effective as a control measure against the Medfly on citrus and stone fruits in South Africa, Spain (Nayem Hassan, pers. comm.), Tunisia (Bouagga 2012), and other countries over the past few years. In these trials, a single application provided a season-long protection.

Table 3. GF-120, Magnet MED, and Ceranock A&K systems juxtaposed (Bouagga 2012).

"A & K" System	Ceranock	Magnet MED	GF-120 NF
Composition	Protein hydrolysate; alpha-cypermethrin	Trimethylamine; Ammonium acetate; Delta-methrin	Ammonium acetate; sugar; protein; spinosad
Field Longevity	4 months	4 months	1 week
Toxicity	No toxicity (no direct contact with crop)	No toxicity (no direct contact with crop)	Toxic for some hymenoptera species and pollinators
Shelf life	2 years if stored in appropriate conditions	2 years if stored in appropriate conditions	2 years if stored in appropriate conditions
Application	By hand	By hand	Conventional pesticide sprayer
Relative Cost	Cheap and competitive	Expensive	More expensive
References	(M.N. Hassan, per. comm.)	(Navarro-Llopis <i>et al.</i> , 2013)	(Vargas <i>et al.</i> , 2001)

2.6.3. Examples of Fruit Fly Attractants

I. Attractants for *C. capitata*:

- Male attractant: TML (tert-butyl esters of 4- and 5-chloro 2-methylcyclohexanecarboxylic acids)
- Female attractant: Mixed amine – Ammonium acetate – ammonium bicarbonate – putrescine – trimethylamine.

(Attractants for *Dacus ciliatus* are mixed amines as for *C. capitata*.)

II. Attractants for *B. zonata*, *B. dorsalis*, *B. invadens* (African invader fruit fly), and *Dacus frontalis* (squash or melon fruit fly):

- Male attractant: ME.
- Female attractant: Protein hydrolysate, trimethylamine, ammonium acetate, putrescine.

III. Attractants for *B. cucurbiata* (Cucurbit fruit fly)

Male attractant: Cuelure [4-(p-Acetoxyphenyl)-butan-2-one].

Female attractant: Protein hydrolysate.

Chapter 3

Materials & Methods

The treatments depended on testing two different parapheromone-mediated techniques for the control of mixed populations of *C. capitata* and *B. zonata* in the same orchard.

The Ceranock A&K technique and Zonatrak MAT were used in apricot and mango orchards, which are two different hosts for both fruit flies. It is worth noting that Zonatrak is obviously also an A&K technique, but it can be termed a MAT due to the fact that such an A&K technique is designed to target only males of Genus *Bactrocera* fruit flies.

3.1. Zonatrak Male Annihilation Technique

Zonatrak is an emulsified wax formulation developed by Russell IPM Ltd. in 2012 (Fig. 17). It targets males of *B. zonata* to significantly reduce male fruit flies. It is an innovative A&K system based on ME and a toxicant, with the aim of combating fruit fly species without directly applying a conventional pesticide. Zonatrak is used for the control of a number of *Bactrocera* fruit flies, including (besides *B. zonata*) the Oriental fruit fly *B. dorsalis* and the African invader fruit fly *B. invadens*.



Active ingredients: • ME • Spinosad (contact biopesticide)

Application rate: 200 dollops/hectare (each dollop being 2 grams).

Method of application. A 5 to 7 mm-wide opening is made at the top of the cartridge and a 2-gram dollop is applied by pressing the cartridge using a caulking gun. Dollops are applied on the trunks or branches in a shady part of trees and bushes at heights of 1–1.5 meter for trees or 0.5 meter for bushes. Only one application is required throughout the season. The application can be manual for small orchards or mechanical for large ones.

3.2. CeraNock A&K Technique

CeraNock for *C. capitata* is a bait station developed by Russell IPM Ltd. in 2011 (Fig. 18). The females of *B. zonata* are also attracted along with the females of *C. capitata*. The technique is based on a fruit fly attractant released from a bait station containing a lethal dose of an insecticide.



Active ingredients:

- Protein Hydrolysate + plant extract → 5g/Bait station
- Alpha-Cypermethrin → 0.01g/Bait station

Inert material (polyurethane and polyethylene) → 10.3g/Bait station

Application rate: 400 bait stations/hectare. Generally, a bait station is placed on each tree, with a 4-meter distance between any 2 stations. Bait stations are also placed in an area 50 square meters wide around the treatment plots. Area-wide application can eradicate a fruit fly population. The number of CeraNock stations can be reduced depending on temperature and insect pressure.

3.3. Monitoring Traps

Monitoring traps are generally applied 6–8 weeks before fruit color break. McPhail traps were used for the monitoring scheme.

3.3.1. McPhail Trap Description

The McPhail trap, also commonly known as the flycatcher, was first developed in Europe as a housefly trap before the turn of the nineteenth century (Steyskal, 1977). In an experiment on the carrion of a dead sparrow, German scientist Friedrich Dahl attempted to quantitatively determine the kinds of insects attracted to the carrion using the first design of such a trap (Dahl, 1896). The trap was later developed by M. McPhail and C.I. Bliss as a protein-baited trap used for the observation of the Mexican fruit fly population in Cuernavaca, Mexico, in 1928 and 1929 (IAEA, 2003).

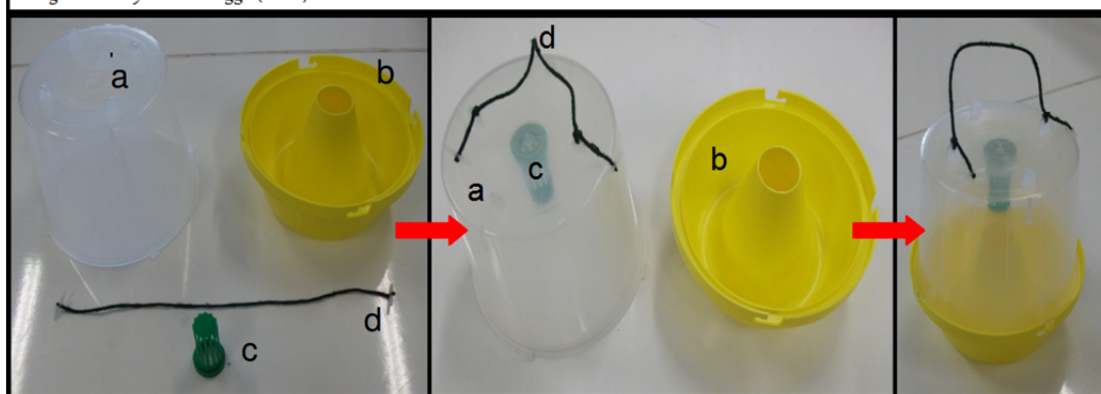
The McPhail traps are now used around the world for the monitoring of fruit flies in area-wide monitoring programs. They are also used in mass-trapping programs with an increased number of traps as compared to those used for monitoring. Further, the traps are used for Sterile Insect Technique programs and in quarantine areas. While the trap is commonly used as a catcher of a range of fruit flies, it has shown a very good performance in attracting *C. capitata* flies, particularly females (Katsoyannos and Papadopouls, 2004).

A reusable season-long trap, the McPhail trap can be used for solid and liquid attractants, and captured insects can be easily seen through the clear plastic cover. It is recommended to hang McPhail traps in the sunny part of the tree at a height of 1.5 to 2 meters with 15 to 20 meters between each 2 traps.

The McPhail traps used for the trials of this study were provided by Russell IPM Ltd. The trap weighs around 150g and consists of the following components (Fig. 20):

1. A yellow bottom section shaped as an inverted funnel serving as an entrance for the flies (90 mm high and 165 mm in diameter).
2. A transparent top section (140 mm high and 70 mm in diameter).
3. A green cage fastened to the inner top of the transparent section to house the DDVP strip.
4. A cord for hanging the trap on trees.

Figure 20. McPhail trap installation
Images courtesy of S. Bouagga (2012)



For the trials of this study, the McPhail traps (which were used for monitoring purposes only) were fed with three different attractants.

3.3.2. TML in McPhail trap

TML is a synthetic insect attractant manufactured for crop protection applications, particularly the control of male Medflies. The name assigned to the compound by the International Union of Pure and Applied Chemistry (IUPAC) is tert-butyl (\pm)-4(or 5)-chloro-2-methylcyclohexanecarboxylate. The name assigned by the Chemical Abstracts Service (CAS) is 1,1-dimethylethyl 4(or 5)-chloro-2-methylcyclohexanecarboxylate (BPDB: Bio-Pesticides Data-Base, 2013).

The 4/5-chloro-isomers in TML serve as strong attractants of male Medflies. Males attracted to the traps baited with TML are retained by an adhesive or killed by an insecticide. While TML is physically a water-white liquid, it is commercially supplied in the form of slow-release formulations, such as plastic bags containing liquid TML isomers or polymer plugs (CAB International, 2009). The latter is the formulation in which TML was used for the trials of this study.

3.3.3. ME in McPhail trap

ME is a naturally occurring substance found in a number of food sources, such as spices and oils. It is also found in nutritionally important foods, such as bananas and oranges (Robison and Barr, 2006).

The CAS name assigned to the compound is 1,2-Dimethoxy-4-(2-propenyl)benzene, while the IUPAC systematic name is 1,2-Dimethoxy-4-prop-2-en-1-yl-benzene. The compound is naturally a colorless to pale yellow liquid with a clove-carnation odor and a bitter taste (WHO, 2012). When used for plant protection purposes, it is also supplied in the form of slow-release formulations, as in the case of TML. The ME used for the trials of this study was contained in slow-release polymer plugs.

3.3.4. Femilure in McPhail trap

Femilure is a commercial product based on a mixture of the following organic amino derivatives: ammonium acetate, putrescine, and trimethylamine. The mixture is designed to attract female Medflies. However, the trials of this work showed that it could also attract flies of other genera, as will be shown in the results section.

According to the manufacturer, the slow-release effect of the product lasts for a period of up to 18 weeks. The product is provided by the manufacturer in the form of compressed paper sheets dipped in the mixture of organic amino derivatives.

3.3.5. DDVP Trapping Strips

Dichlorvos or 2,2-Dichlorovinyl dimethyl phosphate, commercially known as DDVP, is an organophosphorus insecticide used for agricultural and veterinary pest control. It is commercially available as solid strips used in bucket traps to kill attracted flies. The flies attracted to the trap are killed through the slow-release, volatile effect of the solid strips. One strip is enough for each bucket trap, and replacement is recommended every 6 weeks (South African Department of Agriculture, Forestry and Fisheries, 2012).

The strips used for the trials of this study are yellow in color and are impregnated with dichlorvos, with dimensions 27mm x 10mm x 5 mm and weight 1.6mg (Fig. 19).

Figure 19. Trapping strips (DDVP) used in monitoring traps of the trials
Images courtesy of S. Bouagga (2012)



3.4. Data Collection

Trap catches. Trap catches were taken once a week. Trapped insects were counted and recorded separately as male and female trap catches. After the counting, trapped insects were discarded.

Fruit infestation data. The numbers of fallen fruits, fruits with oviposition punctures, and/or infested fruits were recorded over a number of instances of fruit collection from 10 randomly selected trees in each plot's center.

Replacement of bait stations and lures. When and if necessary, Ceranock bait stations should be replaced after 120 days of application, TML plugs every 4 weeks, and Femilure compressed sheets every 90 days or according to the instructions given by the manufacturer.

Data Analysis. Trap catching data of different monitoring traps were statistically analyzed using the "t"-test.

Experimental design. The experimental design was a Randomized Complete Block (RCB) in both the apricot and mango trials.

Other estimations. Daily and weekly mean temperatures were obtained for the whole periods of the trials.

3.5. Biotopes

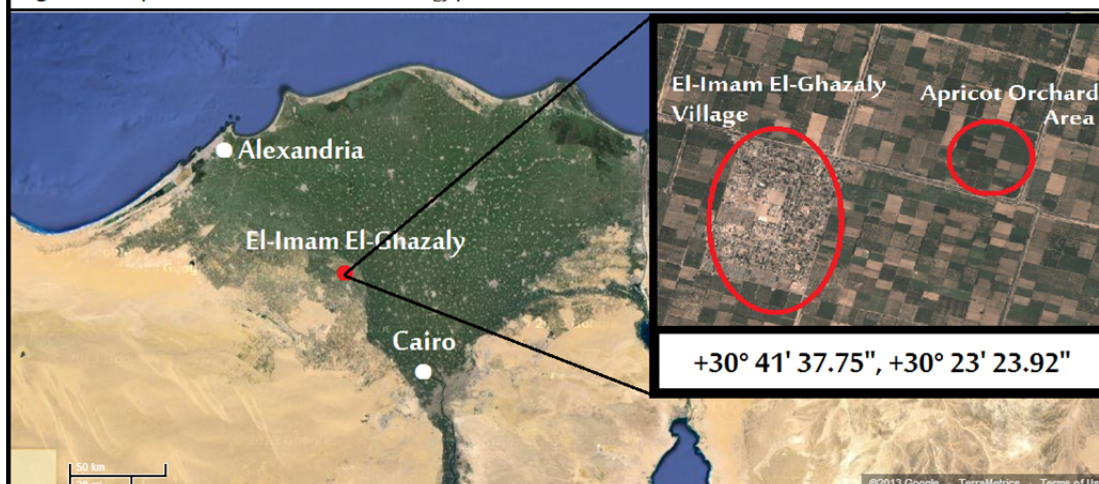
3.5.1. Apricot Trial

The trial was conducted in an apricot orchard northwest of Cairo. The orchard is located in the confines of a village called El-Imam El-Ghazaly in Al-Beheira Governorate of Egypt.

It is also important to mention that the orchard is part of a newly reclaimed desert land, and is surrounded by mango, apricot, and citrus orchards.

The entire village of El-Imam El-Ghazaly along with its confines is only 25 years old, and the agricultural lands in its confines were given to university graduates as part of an Egyptian program for desert land reclamation in the 1980s. The exact location of the orchard is +30° 41' 37.75", +30° 23' 23.92" (Fig. 21).

Figure 21. Apricot trial site in Al-Beheira, Egypt

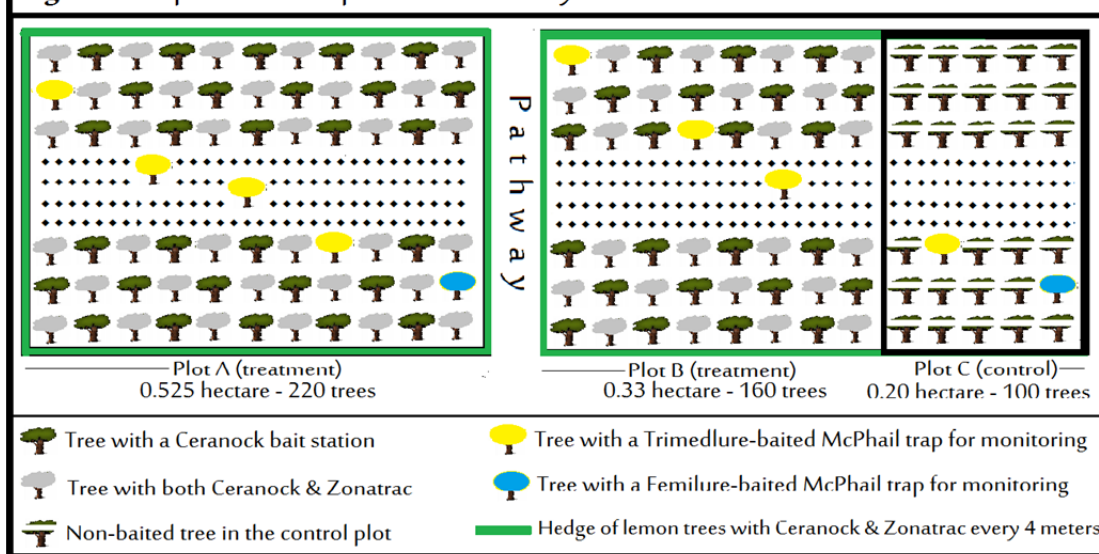


Area, Layout, and Trapping Procedure. The orchard's total area is 1 hectare cultivated with an apricot variety called 'Canino'. The total area was divided into 3 plots: 2 treatment plots and a control plot, as shown in Fig. 22. The control plot was sprayed with an insecticide based on two pyrethroid active ingredients: lambda cyhalothrin and tetramethrin. The insecticide, commercially known as Lambada Plus, is the orchard owner's conventional control measure.

Due to the topography of the orchard (particularly the pathway dividing the orchard into two equal areas), there was no choice but to have 3 plots of unequal area. Thus, the areas for Plots A, B, and C were laid as follows:

- Plot A (treatment): 0.525 hectare with 220 apricot trees
- Plot B (treatment): \approx 0.33 hectare with 160 apricot trees
- Plot C (control): \approx 0.20 hectare with 100 apricot trees

Figure 22. Apricot trial: experimental area layout



In line with the protocols laid out by the manufacturer of the A&K bait station Ceranock and the bait-based MAT Zonatrac (Russell IPM Ltd., 2013; Russell IPM Ltd., 2012), the application rate of Ceranock was 400 bait stations/hectare, and that of Zonatrac was 200 dollops/hectare (each dollop being 2 grams).

For plot A, all trees (220 trees) were baited with Ceranock bait stations, which attract and kill female Medflies and female PFFs. Every second tree (a total of 110 trees) was baited with a 2-gm dollop of the Zonatrac paste, which attracts and kills only PFF males.

Plot B (160 apricot trees) had the same pattern of the treatment applied in Plot A, relative to the number of trees: 160 Ceranock bait stations (1 station/tree) and 160 Zonatrac paste dollops, each dollop being approximately 2 grams (1 dollop/tree).

Apart from the monitoring traps that fell within the area of the control plot (Plot C), there was normally zero application of either Ceranock bait stations or Zonatrac paste dollops. However, the area of that plot (100 trees) was sprayed by the orchard's owner 4 times between the first appearance of fruits and harvest.

As shown in Fig. 22, two types of monitoring trap were applied: TML-baited McPhail traps and Femilure-baited McPhail traps. The trial witnessed an application of a total of 10 McPhail monitoring traps as follows: 8 TML-baited traps + 2 Femilure-baited traps. Plot A had 4 TML-baited traps + 1 Femilure-baited trap, while Plots B & C had 4 TML-baited traps + 1 Femilure-baited trap (Fig. 22).

The periphery of the apricot orchard, which was mainly hedges of lemon trees and sometimes casuarina trees, was baited at 4-meter intervals with Ceranock bait stations and Zonatrac dollops.

After 8 weeks of the trial's onset, "shoot strikes" and wilting terminal leaves characteristic of the damage caused by the overwintering larvae of *Anarsia lineatella* were observed in the orchard. With the aim of not compromising the trial's procedure by the chemical sprays conventionally used by the orchard's owner for such cases, 100 envelopes of *Trichogramma* spp. were provided by the Aswan-based Center for Bio-Organic Agricultural Services and applied in the orchard. Fig. 23 shows the different types of baits and traps used during the apricot trial.

Figure 23. Ceranock bait station (top left), Zonatrax paste dollop (top right), McPhail monitoring trap (bottom left), and *Trichogramma* spp. biocontrol envelope (bottom right) applied in apricot orchard trial.

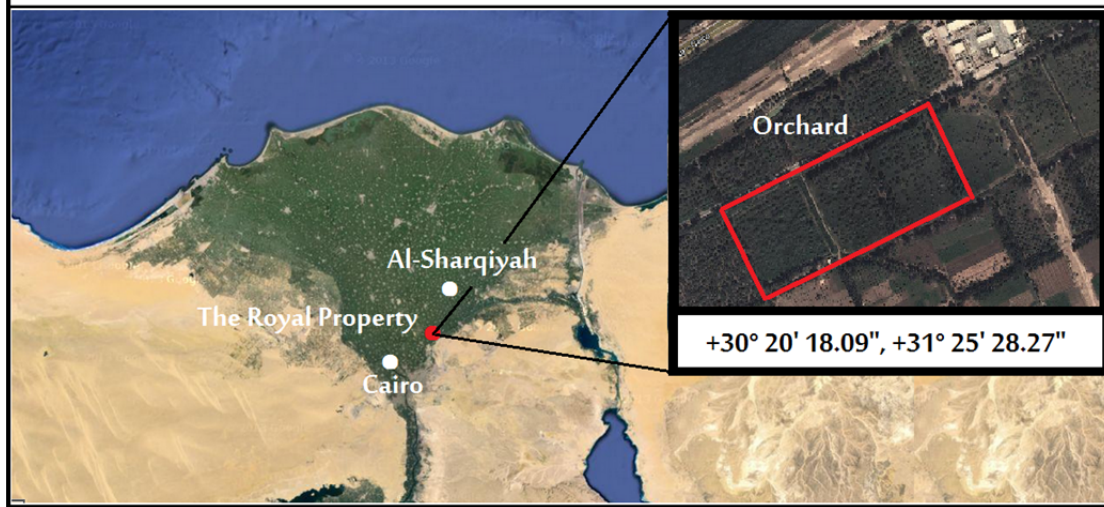


3.5.2. *Mango Trial*

The trial was conducted in 3 mango plots in Al-Sharqiyah Governorate northeast of Cairo. The plots constitute only a small part of a wide agricultural land of more than 1,000 hectares called the Royal Property, which used to be (as the name entails) a property of the Royal Family in Egypt before the 1952 Revolution. Now administered by the Egyptian Ministry of Endowments, this relatively large area is mostly cultivated with mango and citrus and is typically representative of Egypt's so-called "old" clay land.

The trial was conducted on an area of 1.25 hectare cultivated with mainly four mango varieties: 'Owaisi', 'Hindi', 'Dabsha' and 'Baladi'. The exact location of the orchard is +30° 20' 18.09", +31° 25' 28.27" (Fig. 24).

Figure 24. Mango trial site in Al-Sharqiyah, Egypt

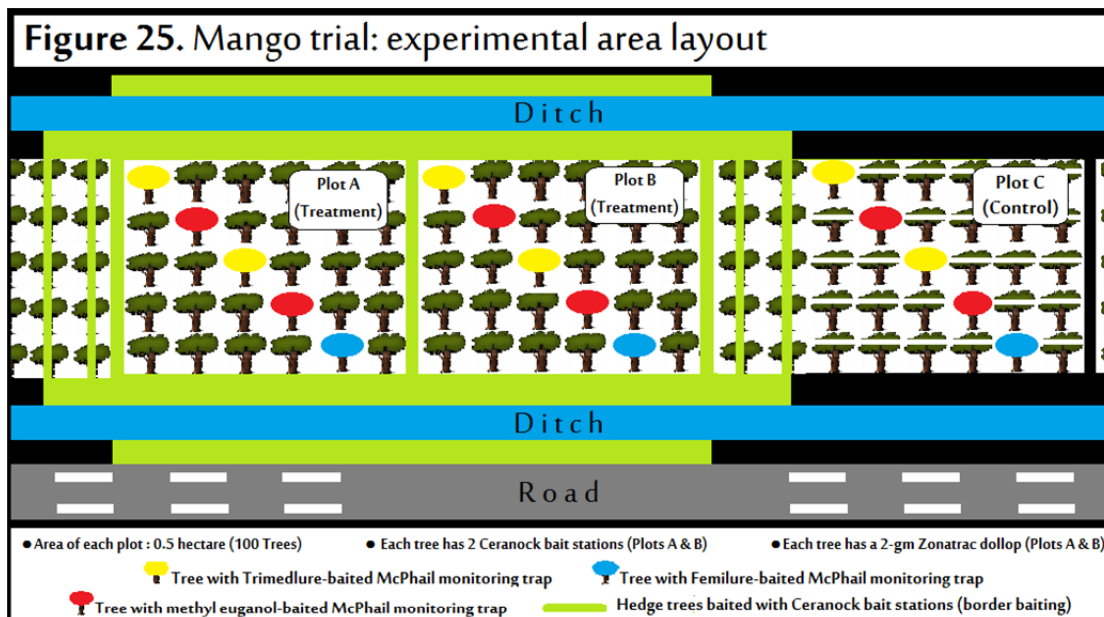


As shown in Fig. 24, the total area is naturally divided into 3 land plots, each being 0.42 hectare in area. Two plots were used as treatment plots where both treatments (Ceranock and Zonatrac) were applied, while the third plot was utilized as a control plot. The control plot normally witnessed zero application of any of the treatments used in the other two plots, and it was also left untreated by any chemical or other control measures (unlike the case of the apricot trial's control plot).

Area, Layout, and Trapping Procedure. Each treatment plot (Plots A & B) had 200 Ceranock bait stations in line with the manufacturer's protocol (Russell IPM Ltd., 2013). As each plot has 100 trees, each tree had 2 Ceranock bait stations. Also, in accordance with the Zonatrac test protocol (Russell IPM Ltd., 2012), each tree had a 2-gm Zonatrac dollop.

Apart from the McPhail traps applied for monitoring purposes, Plot C (control) had zero application of either Ceranock bait stations or Zonatrac paste dollops.

As for the monitoring scheme, a third type of monitoring trap was applied for better monitoring efficiency. ME-baited McPhail traps were used to monitor the population of *B. zonata* in the area. The monitoring scheme was thus laid as follows. Each plot, including the control plot, had 5 McPhail monitoring traps: 2 TML-baited traps + 2 ME-baited traps + 1 Femilure-baited trap (Fig. 25).



As shown in Fig. 25, for a better border defense against Medfly and PFF females, the border areas of the treatment plots were baited with Ceranock bait stations more extensively than in the case of the apricot trial.

Chapter 4

Results & Discussion

4.1. Apricot Trial

4.1.1. Trap Catches

The Ceranock bait stations were applied in Plots A and B on 12 March 2013, which was approximately 6 weeks before fruit color break. The application time was set in accordance with the manufacturer's protocol for application (Russell IPM., 2013). Also, in line with the manufacturer's protocol for Zonatrac application (Russell IPM., 2012), the Zonatrac paste was applied in the two treatment plots on 24 March 2013, which was around the time adult *B. zonata* start their activity.

The data in Table 4 represent the weekly trap catches of the two types of monitoring trap applied (TML- and Femilure-baited traps) over a period of 11 weeks (2nd week of March until 1st week of June 2013). All numbers represent catches of Medflies, either male Medflies in the "TML_n"-labeled columns or female Medflies in the "Femilure_n"-labeled columns.

Table 4. Weekly trap catches recorded in 8 TML-baited McPhail traps and 2 Femilure-baited McPhail traps distributed throughout the apricot treatment and control plots between mid-March and early June 2013 at El-Imam El-Ghazaly Village in Al-Beheira Governorate, Egypt.

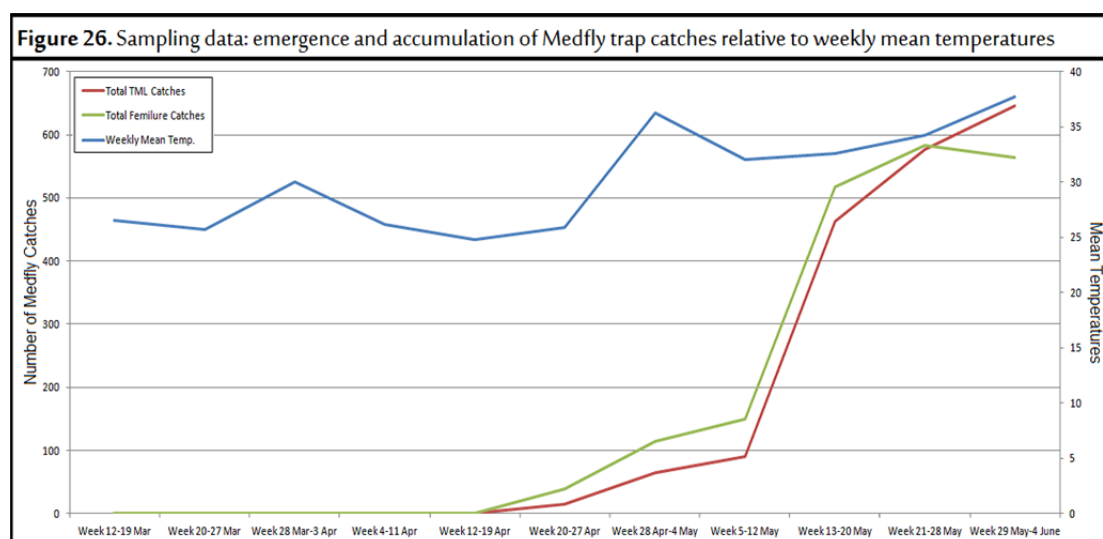
Week	Plot A (Treatment)					Plot B (Treatment)			Plot C (Control)	
	Bait of McPhail Monitoring Trap					Bait of McPhail Monitoring Trap			Bait of McPhail Monitoring Trap	
	TML1	TML2	TML3	TML4	Fem 1	TML5	TML6	TML7	TML8	Fem 2
Week 1 (12-19 March)	0	0	0	0	0	0	0	0	0	0
Week 2 (20-27 March)	0	0	0	0	0	0	0	0	0	0
Week 3 (28 March-3 April)	0	0	0	0	0	0	0	0	0	0
Week 4 (4-11 April)	0	0	0	0	0	0	0	0	0	0
Week 5 (12-19 April)	0	0	0	0	0	0	0	0	0	0
Week 6 (20-27 April)	4	3	6	5	21	7	3	8	4	17
Week 7 (28 April-4 May)	23	13	15	12	47	27	22	29	22	68
Week 8 (5-12 May)	31	21	25	19	53	37	39	45	26	97
Week 9 (13-20 May)	43	49	57	31	216	243	62	74	292	302
Week 10 (21-28 May)	98	89	78	74	249	395	29	82	324	335
Week 11 (29 May-4 June)	124	117	97	83	237	407	43	109	355	327

While both types of monitoring trap (TML and Femilure) had zero Medfly catches up until Week 5, it is worth mentioning that Femilure-baited traps had weekly catches of non-target flies, as will be shown in Section 4.1.2.

As shown in Table 4, Medfly catches started to appear in both types of monitoring trap as of Week 6 between 20 and 27 April 2013, which witnessed

a gradual rise in weekly mean temperature (approximately 26.4°C from 24.7°C in the previous week). In the weeks following Week 6, the catching trend showed an increase in the number of catches relative to a slight, yet fluctuating, increase in daily mean temperatures. The prevailing mean temperatures during the season of apricot in Egypt do not generally contradict the temperature range suitable for the development of Medfly's different life stages: 25 to 26.1°C for shortest larval life (6 to 10 days), 24.4 to 26.1°C for the minimum duration of pupal stage (6 to 13 days), 24.4 to 25.6°C for female flies' readiness to mate (6 to 8 days after eclosion), and 20 to 22.2°C for earliest possible oviposition 4 to 5 days after emergence (Thomas *et al.*, 2001).

The rise in daily mean temperatures is known to contribute to the activity of Medflies in the field, besides biological factors that include age, body size during adult development, and feeding (Peñarrubia-María *et al.*, 2012). In Egypt, seasonal abundance data suggest that milder climate conditions in spring, autumn, and early winter favor the emergence of both the Medfly and PFF (Elnagar *et al.*, 2010). Temperature rise to levels above 33°, however, hinders the activity of Medflies (Sadoud-Ali *et al.*, 2011). The graph in Fig. 26 demonstrates the direct relation between temperature rise and Medfly activity during the trial.

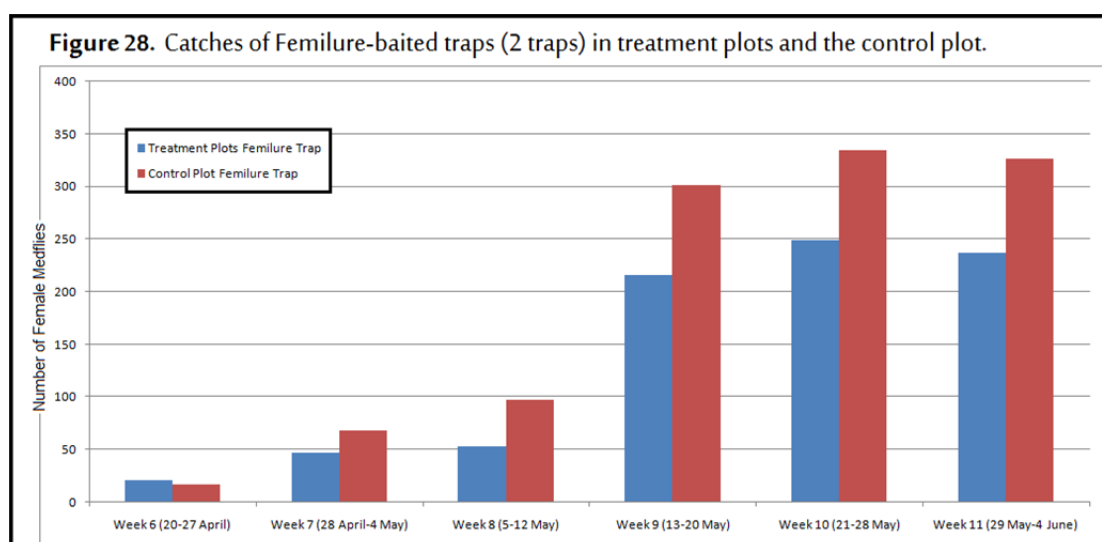
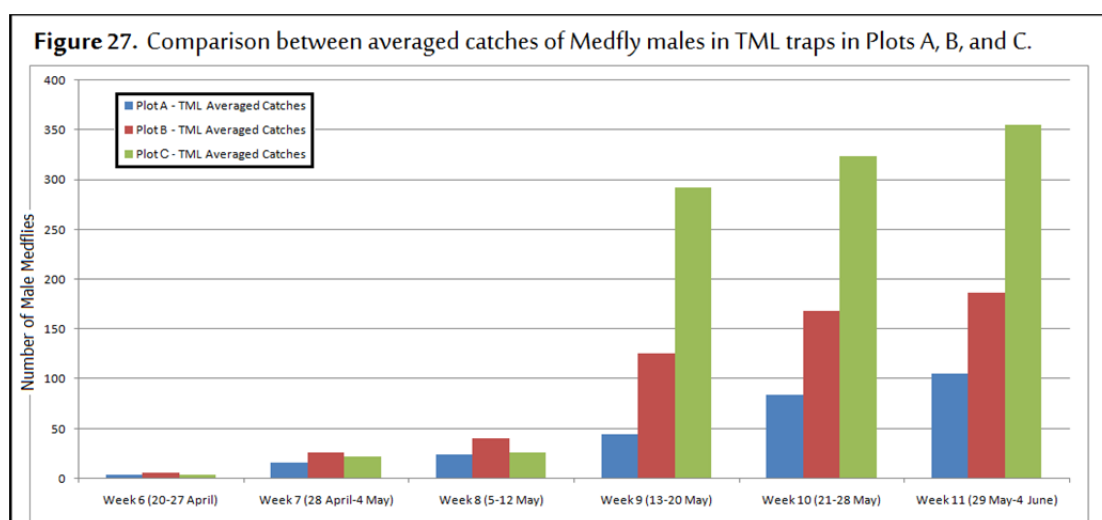


As mentioned in Section 3.6.1, three sides of the apricot orchard were surrounded by mango, apricot, and citrus orchards. In the context of the influence of surrounding fruit crops on Medfly population and infestation rate in different regions of the Italian island of Sardinia, Delrio and Prota (1977) reported that Medfly populations were found to be most abundant in the southern part of the island, where the climate is subtropical and a great variety of fruits are grown. In their survey, Delrio and Prota concluded that peach and

citrus were the most severely attacked fruit species, especially when they are grown together, and that apricot also suffered heavy losses in some years.

The graph in Fig. 27 compares Plots A, B, and C in terms of weekly mean catches in TML traps (male flies). In the last three weeks prior to harvest, TML-baited traps in the control plot had a significantly higher number of catches (approximately 300–350 flies/trap/week) than those recorded in Treatment Plot B, which in turn had higher catches than those in Treatment Plot A.

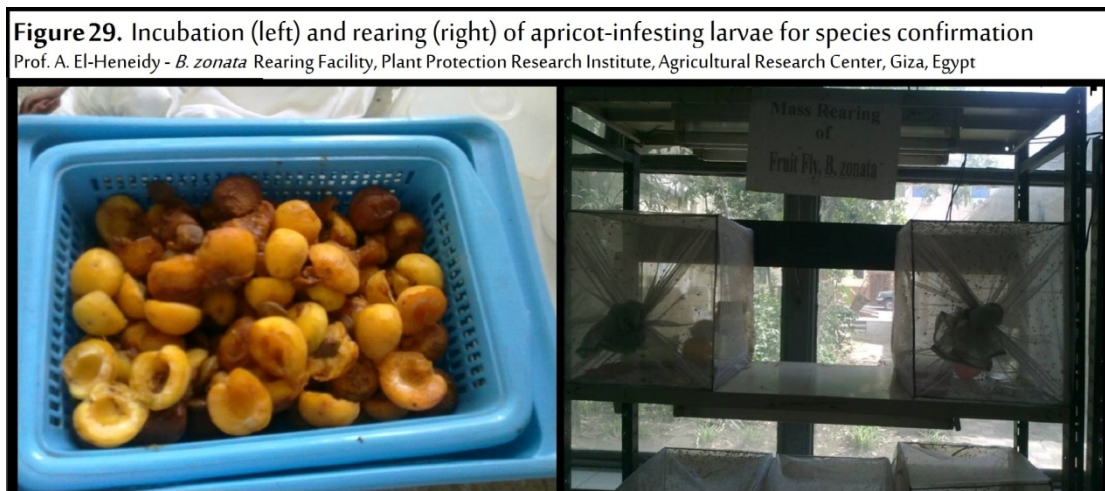
An intuitive explanation is normally the fact that the control plot was void of Ceranock bait stations, which contributed to the higher number of flies therein. This explanation is supported by the fact that the same trend of catching was observed with the 2 Femilure-baited traps. As shown in Fig. 28, as of Week 7 (early May) up until Week 11 (harvest in early June), the Femilure trap of the control plot had higher weekly catches than those in the Femilure trap of the treatment plots, with a weekly difference averaging about 55 flies.



As for the PFF, which was expected to be present in a population mixed with that of the Medfly area-wide, the results based on close inspection of monitoring traps showed the fly population to be zero. The reason why the fly was expected in the area is twofold.

- First, the fly—shortly after it had been identified in significant numbers near Alexandria in 1998—has been considered widespread in Egypt in a number of areas nationwide (EPPO, 2013), including the entire Nile Delta region, at the border of which the apricot trial orchard is located. The fly is even present in very dry areas with few host plants, as well as on isolated trees (ibid). The PFF is also recorded in a city called Al-Dalangat about 50 kilometers away from the apricot trial area in the same governorate (El-Gendy, 2013).
- Second, the PFF is normally recorded in apricot orchards in Egypt and is known to cause severe damage therein, if no proper control measure is applied (Saafan *et al.*, 2005; Elnagar *et al.*, 2010).

The PFF was, however, never found in Femilure-baited monitoring traps, which attract both Medfly and PFF females. As a confirmation procedure that the area had zero PFF population, after the first fruit damage assessment was performed, all dissected apricots (along with the larvae within) were incubated over a soil layer to facilitate pupation. The pupae were then moved to a rearing cage. The process, conducted in a *B. zonata* mass rearing facility at the Egyptian Agricultural Research Center, resulted in a fruit fly population consisting of 100 percent Medflies (Fig. 29).



Non-Target Fly Catches. Back to the non-target flies trapped in Femilure-baited McPhail traps of both treatment and control plots, such flies were captured in significant numbers over the first 5 weeks: around 70 flies/trap/week (Fig. 30). However, as of Week 6, their catching rate started to decrease to around 20 flies/trap/week in parallel to the increase in Medfly

catching rate (Table 4). A sample of these non-target flies was collected and sent to Entomology Professor Salwa Kamal, head of the Entomology Museum at the Ain Shams University Faculty of Science. All non-target flies were identified as flies of Genus *Muscina* (Diptera: Muscidae) as confirmed by the taxonomic procedures (pers. comm.).

Figure 30. Flies of Genus *Muscina* attracted by Femilure. Image taken in field on 24 March 2013



4.1.2. Apricot Fruit Damage Assessment

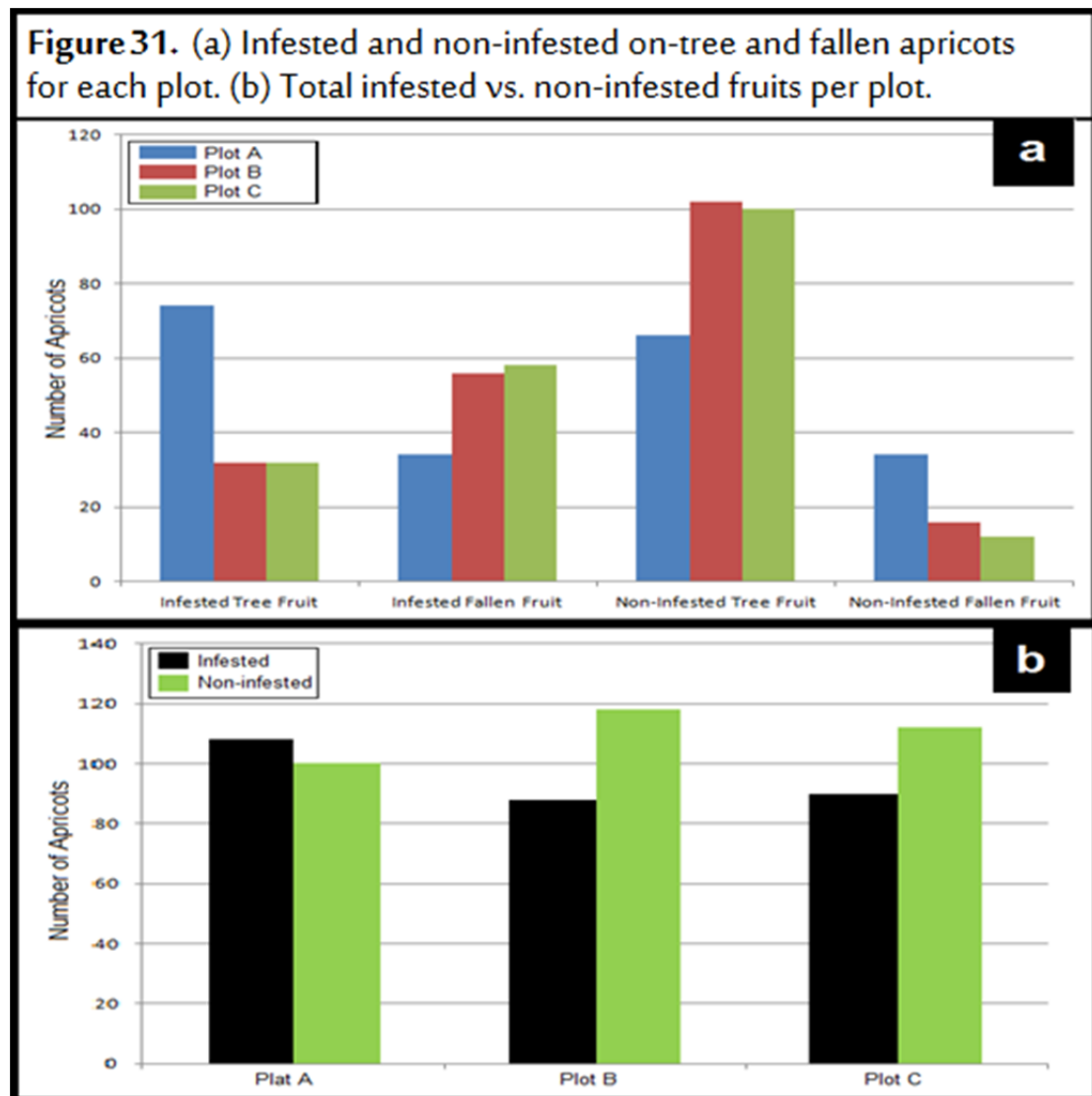
On the 22nd and 29th of May, 77 days of the trial's onset, a total of 600 apricots were collected for fruit damage assessment. In Table 5, numbers of infested and non-infested apricots in each plot are laid out, with a distinction between fruits collected from trees and fallen fruits.

Table 5. Infested and non-infested fruits (400 apricots) collected from the trees and orchard floor of Plots A, B, and C.

	Infested Tree Fruit	Infested Fallen Fruit	Non-Infested Tree Fruit	Non-Infested Fallen Fruit	Infestation (%)
Plot A	74	34	66	34	51.92%
Plot B	32	56	102	16	42.72%
Plot C	32	58	100	12	44.55%

Out of 200 apricots for each plot, Plot A had the highest number of infested fruits on trees (35.58% of its total sample), while Plots B and C had an equal number of on-tree infested apricots (15.53% and 15.84% of their total samples, respectively). However, the number of infested fallen fruits in Plot A was the lowest (16.35% of its total sample) compared to those in Plots B and C (27.18% and 28.71% of their total samples, respectively). All in all, Plot A had the highest infestation rate (51.92%), followed by Plot C (44.55%) and Plot B (42.72%).

The graph in Fig. 31 (a) shows the results of fruit damage assessment in terms of the numbers of infested and non-infested apricots collected from the trees and orchard floor of each plot. Fig. 31 (b) draws a comparison between total infested versus non-infested fruits in each plot.



A post-trial evaluation of the reasons behind such a relatively high infestation rate in all plots resulted in a number of hypotheses.

First, while the manufacturer's protocol for Ceranock application (Russell IPM, 2013) recommends the treatment area to be at least 1 hectare, the total area of Treatment Plots A and B did not exceed 0.86 hectare.

Second, one of the manufacturer's recommendations (ibid) was to deploy bait stations in a peripheral area 50 square meters deep around the treatment plots, with 4 meters between any two stations. However, only a single-row hedge around the treatment plots was baited, due to the limited number of stations shipped from the manufacturer's headquarters in the UK to Egypt. Therefore, any attempt of applying more peripheral baits would have been at

the expense of reducing the application rate inside the treatment plots, and would have also been difficult due to the fact the apricot orchard is located amid an area of small fragmented agricultural lands owned by different farmers.

Third, the Medfly population density in the area was obviously much higher than the baiting system's capacity for "attracting and killing." As apparent from Table 4, the Medfly population witnessed a soaring increase as manifested by the catching rates ranging between 350 and 400 flies/trap/week in some monitoring traps. Such a "booming" population load took place toward harvest at a time when apricots become most vulnerable to female oviposition.

This Medfly population "overload" in the experimental plots was most probably due to an insecticidal repelling effect from neighboring orchards cultivated with insecticide-treated Medfly hosts at three of the apricot orchard's four "frontiers" (see Section 3.6.1). Such a repelling effect—e.g., from malathion (IAEA, 2003)—is thought to have caused the flies to "migrate" in overwhelming numbers to the non-chemically treated experimental plots, in such a way that rendered the application rate less effective. Such a migration is supposed to have been even more "exacerbated" by the attractant odor, which emanated from the treatment plots.

A final note supporting the validity of the previous explanation is the control plot's high infestation rate (44.55%), even though the plot was chemically sprayed 4 times between the first appearance of fruits and harvest.

4.2. Mango Trial

4.2.1. Trap Catches

The Zonatrax paste dollops were applied in Plots A and B on 14 May 2013 at a time when adult *B. zonata* flight was observed in the field, in line with the manufacturer's protocol for application (Russell IPM., 2012). Also, in accordance with the manufacturer's protocol for Ceranock application (Russell IPM., 2013), the Ceranock bait stations were applied in the two treatment plots on 8 June 2013, which is about 6 weeks prior to fruit color break.

As mentioned in Chapter 3 on materials and methods, the monitoring scheme for the mango trial was modified by adding a third type of monitoring trap aimed at enhancing PFF population monitoring (2 traps baited with ME in each plot).

The data in Table 6 represent the weekly trap catches of the three types of monitoring trap applied (TML-, ME-, and Femilure-baited traps) over a period of 13 weeks between mid-May and the last third of August (harvest date).

Numbers in TML_n columns represent male Medfly catches, while those in ME_n columns represent male PFF catches. Numbers in Femilure_n columns are either female Medflies, female PFFs, or both.

Table 6. Weekly trap catches recorded in 3 Femilure-baited McPhail traps, 6 ME-baited McPhail traps, and 6 TML-baited McPhail traps evenly distributed throughout the mango treatment and control plots at the Royal Property orchards in Al-Sharqiyah Governorate, Egypt, between mid-May and late August 2013.

	Plot A (Treatment)					Plot B (Treatment)					Plot C (Control)				
	Femilure 1	ME1	ME2	TML1	TML2	Femilure 2	ME3	ME4	TML3	TML4	Femilure 3	ME5	ME6	TML5	TML6
Week 1 (14-21 May)	0	2	0	0	0	0	5	0	0	0	0	32	42	0	0
Week 2 (22-29 May)	0	2	1	0	0	0	0	0	0	0	2 PFF	10	28	0	0
Week 3 (30 May-5 June)	0	1	3	0	0	0	2	0	2	0	0	11	13	0	0
Week 4 (6-13 June)	0	2	8	0	0	0	1	1	1	0	0	10	15	0	0
Week 5 (14-21 June)	1 PFF	0	7	0	0	0	0	0	0	0	0	6	5	0	0
Week 6 (22-29 June)	0	0	1	0	0	0	2	1	0	0	0	9	32	0	0
Week 7 (30 June-6 July)	0	1	2	0	0	0	4	2	0	0	0	15	43	0	0
Week 8 (7 July-14 July)	0	0	18	1	0	2 MFF+1 PFF	7	12	1	0	0	28	61	0	0
Week 9 (15 July-22 July)	1 PFF	1	33	2	0	2 MFF	15	18	2	0	1 MFF	47	113	0	0
Week 10 (23 July-30 July)	0	1	17	0	1	0	9	18	1	0	2 PFF+1 MFF	39	47	0	0
Week 11 (31 July-6 Aug)	0	1	29	0	2	0	16	22	1	0	4 PFF+1 MFF	62	78	1	1
Week 12 (7 Aug-14 Aug)	0	2	21	1	2	0	7	13	0	0	1 MFF	20	81	0	0
Week 13 (15 Aug-22 Aug)	0	0	24	0	1	0	5	9	0	0	1 PFF	26	95	0	0

As shown in Table 6, the earliest trap catches were male *B. zonata* flies in ME-baited McPhail traps, which had the flies trapped as of Week 1 in mid-May. The TML-baited traps generally had very small numbers of weekly male Medfly catches; the numbers never exceeded 2 flies/trap/week, and the first such catches started as of Week 4. Femilure-baited traps, capable of attracting both PFF and Medfly females, also had very small numbers of both species; numbers never exceeded 5 flies/trap/week.

As with the case of apricot, Femilure-baited McPhail traps also had non-target catches of flies belonging to Family Muscidae, though the average numbers were only 10 flies/trap/week; such non-target catches stopped after Week 4 (mid-June).

The catches in different McPhail traps were enough proof that both the Medfly and PFF existed in mixed populations, though the PFF population clearly outnumbered the Medfly population, with a ratio of about 50 PFFs for each Medly. This comes in line with the findings asserted in the literature about the total or partial displacement of Medfly populations caused by the presence of PFF populations in the same area (El-Heneidy, 2012; EPPO, 2005).

In Egypt, Hashem *et al.* (2001) reported that *C. capitata* has become more restricted in horticultural areas due to the spread of *B. zonata*. Mohamed (2004) found that fruits infested by both flies were mostly infested by *B. zonata* regardless of which fly infested the fruit first. Apricot orchards in Al-

Fayoum Governorate south of Cairo had Medfly populations at much lower levels than those of *B. zonata* (Saafan *et al.*, 2005).

The same phenomenon of displacement caused by *B. zonata* to other tephritid flies, including *C. capitata*, was observed in other countries: India (Kapoor and Grewal, 1986), Mauritius (Seewooruthun *et al.*, 1998), and Réunion (Duyck *et al.*, 2006).

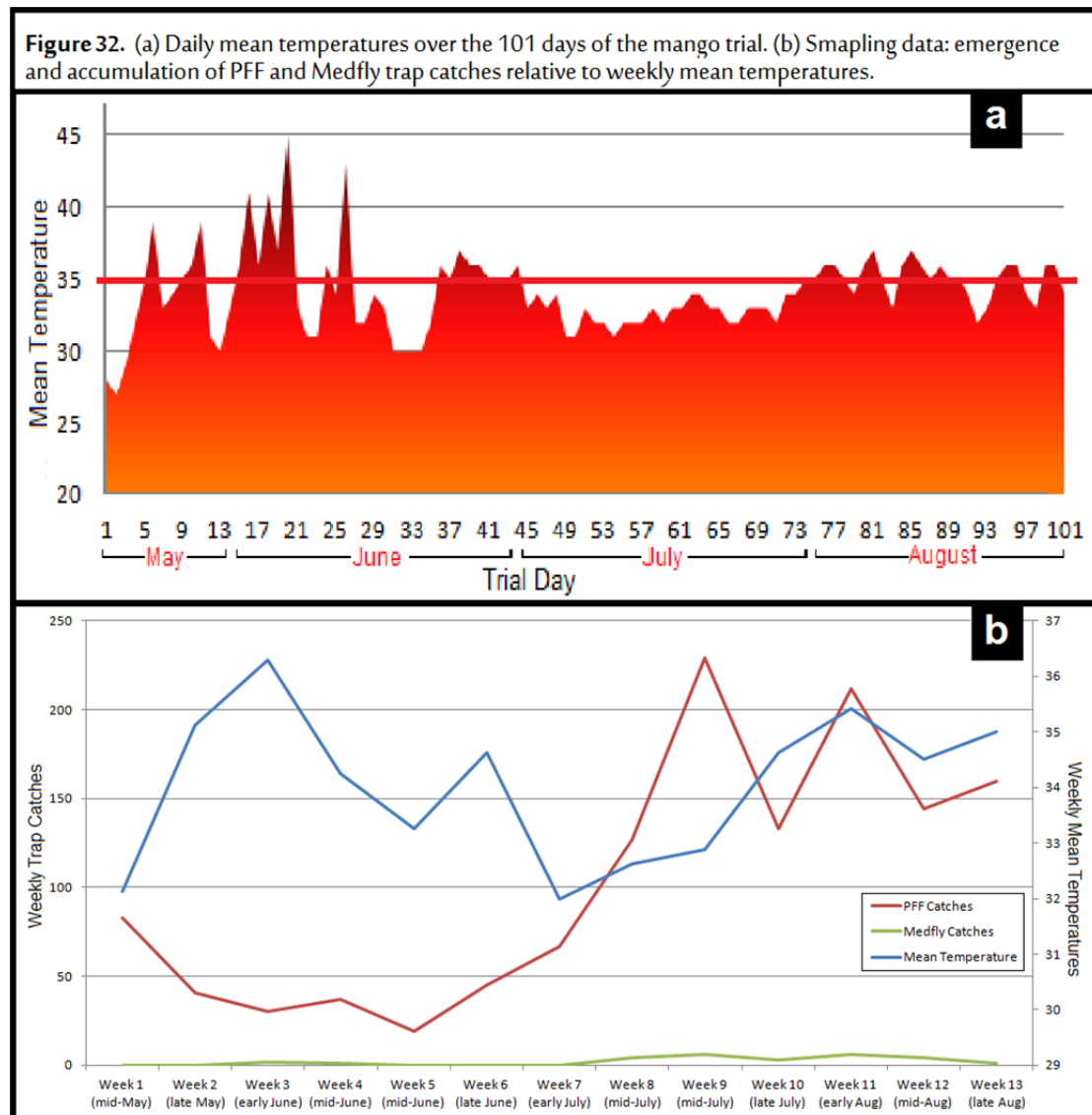
One explanation for such phenomenon is competition for food (CAB International, 2011). Another explanation employs the “r–K gradient” defining the varying invasive capacities of different species. Adaptive strategies aiming at the survival of a species (e.g., fertility and mortality rates, age at first reproduction, and life expectancy at birth) have extremes “r” and “K” between which most species fall (Lévêque, 2003). Species of Genus *Bactrocera* are characterized by type K-demographic strategy traits, which enable them to adapt for competition in saturated habitats more than r-selected species such as *C. capitata*, and the converse is never true (Duyck *et al.*, 2004).

A third explanation referred to by Elnagar *et al.* (2010) is the possible effect of climate change, which has given rise to longer and higher summer temperatures as documented by meteorological records of the Central Laboratory for Agricultural Climate in Egypt. The more hot climatic conditions over the years may have contributed to suppressing Medfly populations whose activity is hindered at temperatures above 33°C (Sadoud-Ali *et al.*, 2011), whereas PFF populations can withstand higher temperatures (Qureshi *et al.*, 1993). This comes in line with results reported from Iraq showing a substantial decrease in Medfly population density at mean temperatures recorded at 46–51°C in August 2010 (Khalaf *et al.*, 2012).

As mentioned in Section 2.4.3, the PFF thrives best at an optimum temperature between 25 and 30°C. However, according to Qureshi *et al.* (1993), 25°C is a key degree of ambient temperature at which a number of life stages develop at their best: egg development, egg hatching, larval and pupal survival, female fecundity, and male/female longevity. On the other hand, 35°C is a degree at which egg hatching completely stops, larval development halts, and pupal development decreases (ibid).

Out of 101 days since the onset of the trial until harvest, 42 days (mostly in June and August) had daily mean temperatures greater than or equal to 35°C—Fig. 32 (a). While this should have contributed to a reduction in Medfly population, it may have also done the same to the PFF population in the area. Still, the reduction in Medfly populations must have been higher under such climatic conditions, which partially explains the PFF-Medfly ratio encountered during the trial. The catching rates of both the PFF and Medfly in relation to

different weekly mean temperatures over the period of the trial are shown in Fig. 32 (b).

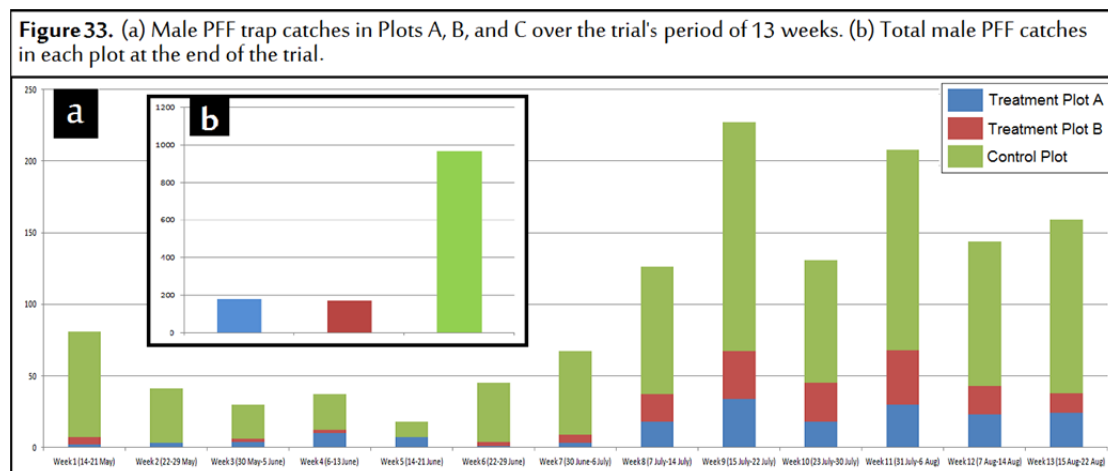


On the other hand, also over the trial's period of 13 weeks, the catching rate of male PFFs per trap was also much higher than that of female PFFs, with a ratio of 101 males for each female.

As there has been a clear explanation for the small Medfly population in the face of the PFF population, the small numbers of trapped female PFFs are explained by the fact that the protein hydrolysate in Ceranock bait stations is also a strong attractant of female PFFs (Gopaul and Price, 2000; EPPO, 2005). Such bait stations were applied in sufficient numbers in treatment plots and along their borders.

It therefore follows that the total numbers of captured male Medflies (20), female Medflies (7), and female PFFs (13) can be neglected. A comparison

among male PFF catches in ME-baited McPhail traps of Plots A, B, and C (2 traps/plot) is drawn in Fig. 33.



As shown in the graph, Plot C (control) had the highest number of male PFF catches throughout the trial as of the first week in mid-May up until harvest in late August (average 74.5 flies/week). Treatment Plots A and B had catching rates apparently lower (averages 13.5 and 13 flies/week, respectively). In sum, at the end of the trial's 13 weeks, Plot C had a total of 968 male PFFs (73.67% of total PFF catches), while the totals for Plots A and B were 177 and 169 flies, which are 13.47% and 12.86% of total catches, respectively.

Such discrepancy between male PFF populations in treatment plots and that in the control plot is normally due to the treatment applied. As a MAT targeting only male PFFs, the Zonatrac paste clearly resulted in a disturbed male-female ratio due to male population reduction in Treatment Plots A and B. Such a minimized male population has led in turn to significant numbers of unmated females, and eventually very few progeny resulted (Mirani, 2007; Ghanim *et al.*, 2010).

Such a few progeny gets even fewer when adult flies become too old to reproduce, as revealed by a demographic analysis in a PFF mass rearing facility, where it was observed that reproduction witnesses a drastic decline in flies older than 35 days (Hussain, 1997).

4.2.2. Mango Fruit Damage Assessment

Sample fruits were collected 4 times in 4 different weeks of the trial: on 19 June (Week 5), 11 July (Week 7), 2 August (Week 11), and 18 August (Week 13). Each fruit collection of the first three weeks included 90 fruits (30 out of each plot), and the last fruit collection during harvest had 180 fruits (60 out of each plot). In total, 450 fruits were collected over the trial's period.

The first 3 collections (up to 2 August) had zero fallen fruits and zero infestation in either the treatment plots or the control plot, possibly due to the thickness of fruit pericarp at that time of the season, which makes female oviposition difficult. Studies conducted at the Pakistani Nuclear Institute for Agriculture and Biology on fruit fly preference for mango varieties, their colors, and pericarp hardness showed that female PFFs prefer to lay eggs in ripened and fully ripened mangoes, and that tough fruit pericarp is less attractive to *B. zonata* flies due to its low total soluble solids (Akhtar *et al.*, 2012).

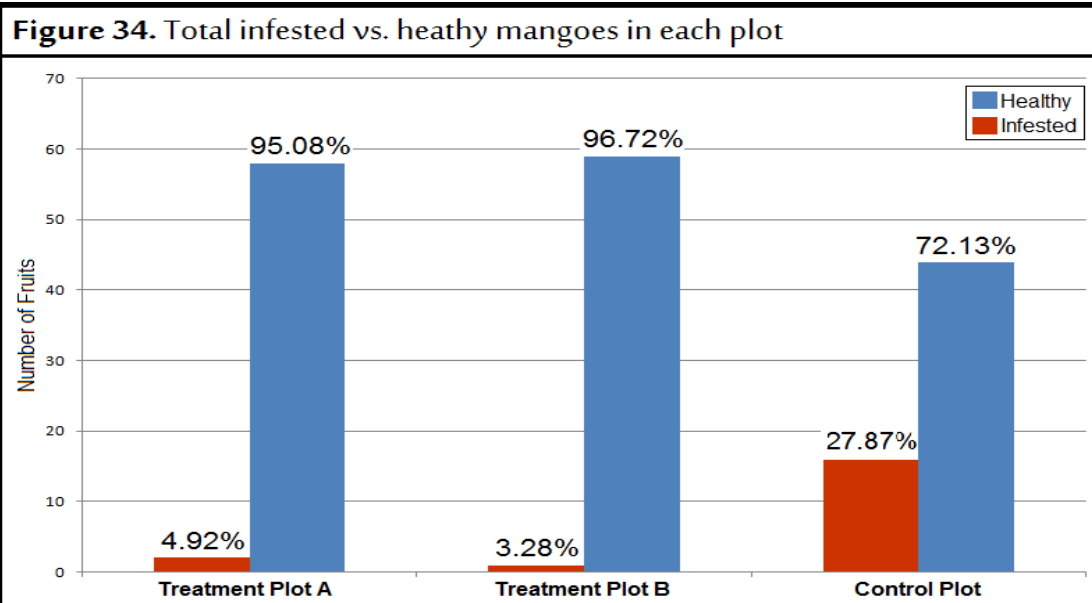
Table 7 shows the results of fruit damage assessment for the last fruit collection. Numbers of infested and non-infested mangoes in each plot are laid out.

Table 7. Infested and non-infested fruits (180 mangoes) collected from Plots A, B, and C.

	Infested Tree Fruit	Infested Fallen Fruit	Non-Infested Tree Fruit	Non-Infested Fallen Fruit	Infestation (%)
Plot A (Treatment)	3	0	58	0	4.92%
Plot B (Treatment)	2	0	59	0	3.28%
Plot C (Control)	7	10	42	2	27.87%

Out of 60 mangoes collected from Plot A, only three fruits were found to be infested. The same number of samples collected from Plot B had two infested fruit. While Plots A and B had zero fallen fruits, the floor orchard of Plot C (control) had quite a significant number of fallen fruits (mostly of the yellow-colored variety 'Baladi' preferred by *B. zonata*). The control plot, which had no control measures whatsoever, normally had the highest infestation rate. Infestation rates for Plots A, B, and C were 4.92%, 3.28%, and 27.87%, respectively.

The graph in Fig. 34 shows the results of fruit damage assessment in terms of infested versus non-infested collected mangoes in each plot.



4.3. Statistical Analysis: “t”-test

The “t”-test was used to determine if the catches of TML-baited traps in the apricot trial’s treatment plots were significantly different from those of the control plot’s TML-baited trap. To create two sets of data suitable for “t”-test data processing, a data transformation of TML-baited trap catches in the treatment plots was performed by taking the means of all TML trap catches. The resulting data column was then used in the “t”-test against the data column of the control plot’s TML trap.

The means for both columns (treatment mean TML trap catches and control TML trap catches) were calculated. Then S^2 was calculated for each column based on the following formula of the “t”-test:

$$S^2 = \frac{\sum(x - M)^2}{n - 1},$$

where x is the individual column score, M is the mean, and n is the number of scores in each column.

The t statistic was then calculated as follows:

$$t = \frac{M_{\text{treatment}} - M_{\text{control}}}{\sqrt{\frac{S^2_{\text{treatment}}}{n_{\text{treatment}}} + \frac{S^2_{\text{control}}}{n_{\text{control}}}}} = \frac{36.07792208 - 93}{\sqrt{251.1318603 + 2020.927273}} = -1.194183126$$

The “t”-test was similarly run on the trap catches of the treatment plots’ Femilure trap and the control plot’s Femilure trap, and the t statistic value was -0.549595593 .

Both t values for the TML and Femilure trap catches in treatment plots against those of the control plot showed the difference between means to be insignificant, as the t statistic value was lower than the t critical two-tail value, with p -values greater than 0.05 (Table 8).

Following the same steps, the “ t ”-test was run on mean ME trap catches of the mango trial’s treatment plots against those of the control plot. The test resulted in a t statistic value higher than the t critical two-tail value, with a p -value less than both the two commonly used significance levels 0.05 and 0.01 (Table 8). This indicates a highly significant difference between mean ME trap catches in the control plot and those in treatment plots, which supports the validity of the results indicated in Section 4.2.

Table 8. Results of the “ t ”-test on mean catches of different traps in treatment plots against those in control plots of the apricot and mango trials.

Trial	Type of Trap	t statistic	t critical (two-tailed)	p-value	Difference
Apricot	TML	-1.194183126	2.085963441	0.246377973	Insignificant
Apricot	Femilure	-0.549595593	2.085963441	0.588681887	Insignificant
Mango	ME	-4.539504706	2.063898547	0.000133946	Significant

Chapter 5

Conclusions

In this study, an A&K technique and bait-based MAT were critically evaluated in terms of their ability to control two key insect pests, *Bactrocera zonata* and *Ceratitis capitata*, supposedly present in mixed populations on apricot and mango in Egypt.

The two techniques were tested together in a sand-soil apricot orchard northwest of Cairo and in a clay-soil mango orchard northeast of Cairo. In the process, the apricot trial site turned out to be void of PFF population, as confirmed by weekly trap catches and later by a mass rearing of the larvae found in infested apricots. However, the mango area had both flies in a mixed population, though the PFF population was obviously predominant over that of Medflies.

The apricot trial witnessed a relatively high infestation rate in both treatment plots, as well as the control plot, due a shortage in border Ceranock bait stations applied, in the face of an overwhelming Medfly population surpassing the application rate capacity.

In the mango trial, both techniques (particularly the Zonatrak MAT) have proved to be successful in substantially reducing infestation on mango, where fruit damage assessment revealed that a 95.08% control and a 96.72% control were achieved in the trial's two treatment plots. On the other side, the percentage of healthy fruit in the control plot mounted to only 72.13%, which corresponds to a loss of more than a quarter of the crop.

Such a low infestation rate in treatment plots was attained through the Zonatrak MAT's ability to achieve a substantial reduction in male PFF population, where the treatment plots had only 13.47% and 12.86% of total male PFF catches area-wide, as opposed to 73.67% of total male catches in the control plot.

While the use of the Ceranock A&K technique was not very successful with apricots, apparently due to the relatively small area of the experimental plots (see results analysis in Section 4.1.2.), it proved to be effective in achieving protection against the mango orchard's female PFF population, in such a way that resulted in only 13 female PFFs attracted to monitoring traps area-wide throughout the trial's period. Although the Medfly population levels were already low due to PFF invasive dominance, the Ceranock technique is still believed to have been effective against female Medflies in the area, based on the very small number of females (7 flies) attracted to monitoring traps throughout the trial's period of 13 weeks.

In a detailed review of the impact of pesticide use on human health and the environment in Egypt, Prof. Sameeh Mansour of the Egyptian National Research Center documented a long list that included human poisoning, toxicity to farm animals, insect pest resistance, destruction of beneficial parasites and predators, food contamination, and pollution of environmental ecosystems. This harm to human, animal, and plant lives has been inflicted by the use of more than 1 million metric tons of pesticides in Egypt's agricultural sector over the past 50 years (Mansour, 2008).

The results of the present study refer to the possibility of adopting the experimented A&K technique and bait-based MAT on a larger scale as control measures alternative to pesticide sprays conventionally used against the PFF and Medfly, which attack together a number of economically important fruit crops in Egypt.

Advantages of the two techniques, besides their eco-friendly characteristics, include a relatively long lifetime in the field (up to 4 months for Ceranock and 3 months for Zonatrak), which results in a reduction in labor required for application. For example, all bait stations and paste dollops for the apricot trial were applied by one person in an area of 1 hectare within two working days. This can be regarded as a significant reduction in labor when compared with the labor needed for pesticide application using a motor-driven sprayer, particularly if we note that the control plot was sprayed 4 times during the apricot season as a habitual control measure followed by the orchard's owner.

Ceranock and Zonatrak controls against mixed populations of the PFF and Medfly can be maximized with the help of field sanitation as a cultural control measure, where infested fruits are continually buried and ripe fruits are harvested on an "as and when needed" basis.

As a first study conducted in Egypt using the two techniques on two different crops, this work can provide a new base for the parapheromone-mediated control of these two fruit flies, particularly in organic farms as a springboard for further spread in small fragmented agricultural lands where pesticides are commonly used for pest control.

However, there is a need for further studies on a larger scale and on other fruit crops under Egyptian field conditions for at least two seasons, with a view to a better estimation of application rates based on population densities of the concerned fruit flies. Successful trials conducted under monitoring from local authorities at the Egyptian Ministry of Agriculture and Land Reclamation can generally help promote the trend of parapheromone-mediated control as a replacement of pesticide treatments.

Chapter 6

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