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Pheromone traps for the assessment of insecticides efficiency and monitoring insecticides resistance in insect field strains

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Abstract:

Up to date insecticides are the backbone of agricultural industry (Agricultural Mass Production) and for public health protection purposes. Annually, trillions of US $ are lost as a result of insect infestations and damages in different agric. Crops around the world , even under the heavy use of insecticidal applications because of the control failure of many inefficient insecticides , that is happen because many insect field strains have developed resistance against these compounds. For the purposes of assaying insecticides efficiency and resistance in insect field strains, it's required to apply huge amounts of insecticides in many sequential treatments, investigate huge amounts of agric. crop fields, collect huge amounts of crop samples (Vegetation and / or fruit parts), rather than environmental pollution, human health hazards, using application equipments which require a lot amounts of effort, time and money. It's recommended here to use pheromone traps technique for the assessment of insecticides efficiency, monitoring insecticides resistance in insect field strains instead of the insecticides conventional applications to overcome all the above mentioned problems and to apply an advanced insecticides resistance management throughout an integrated pest management (IPM) with a very simple, easy, quick, and efficient technique.

Introduction

Formulated pesticides are used in a large scale through the world as a major mean for pest management and control. Although pesticides provide numerous benefits in terms of increased agricultural production and improve its quality, but their efficacy may be not often good because of the development of insecticide resistance in many pest species. Resistance to one or more pesticides has been documented in more than 447 species of insects and mites (Roush and McKenzie, 1987). Pesticide resistance is an increasingly urgent worldwide problem. Resistance in vectors of human disease, particularly malaria-transmitting mosquitoes, is a serious threat to public health in many nations. Agricultural productivity is jeopardized because widespread resistance in crop and livestock pests. Serious resistance problem is also evident in pests of the urban environment, most notably cockroaches.

Resistance to insecticides is one of the most serious problems facing agriculture today. Many previous studies revealed the high resistance of pink bollworm to insecticides in the cotton fields. In Egypt, several

pyrethroids and organophosphorus have been widely used against cotton pests. However, although pyrethroid and organophosphorus insecticides were the most efficient and widely used against bollworms but the onset of resistance development to these compounds in bollworms have been recently documented (Georghiou, 1983; Haynes *et al*., 1987; Miller, 1990 and Shekeban, 2000). The major economic losses due to the pesticide resistance in the USA were: $1.5 billion (Pimentel, 2005). The price of insecticide resistance in lost yields and higher insect control costs is staggering - in some years more than 1 billion $ in cotton for the budworm/bollworm complex alone. In IPM, pheromones are considered to be an essential component because they are used for detecting the economic threshold levels of pest populations and for mating disruption as a direct pest suppression measure. Pheromones of major pests have been found to be effective, economic and eco-friendly in agro-ecosystems in which cotton is cultivated (Tamhankar *et al*., 2000). For resistance management tactics to be effective, resistance must be detected in its early stage (Roush and Miller, 1986) and early detection necessitates using one or more techniques that being accurate, easy, rapid and inexpensive, which would aid production, consultants and extension personal in making informed decisions on adequate control measures (Mink and Boethel, 1992). Firstly, the traditional approach uses complete dose-response tests with 4-5 doses that produce 10-90% mortality. Resistance is expressed in terms of the ratio of LD_{50} or LC_{50} of the resistant strain to that of the susceptible strain. Alternatively, an approach called the discriminating or diagnostic dose test was used where one dose is often investigated and the mortality of the susceptible test strains is compared (Pasquier and Charmillot, 2003).

Another approach is the attracticide method which was developed in summer of 1985 as a rapid test to determined resistance in pink bollworm adult in cotton fields. The attracticide method was full implementation in 1986 and 1987 as an effective method to monitor insecticide resistance to pink bollworm to a wide range of insecticides (Miller, 1986 and Haynes *et al* ,1986 c and 1987). Detection of changes in response is needed, especially in the early stages of resistance development. Monitoring insecticide resistance in field population moth is in great importance in resistance management programs (Tabashnik and Cushing, 1987). In Egypt, very few dose mortality studies using attrcticides resistance monitoring technique were carried out (Al-Beltagy *et al*., 1993). Therefore, it is important to study efficacy of insecticides against insect field strains in Nile Basin countries to establish a program for controlling and reducing the resistance level of these pest to insecticides. Such a program could be efficiently used to reduce number of insecticide sprays; cost of insecticides and increases the crop production per unit.

This review is concentrated on the use of attracticide resistance monitoring technique against different insect field strains and its modified procedure.

1. Attracticide resistance monitoring technique (ARMT) review:

Henneberry and Clayton (1982) conducted studies to determine the relationship between catches of male insects in pheromone-baited traps and crop infestation. Numbers of male moths of the pink bollworm, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelechiidae), caught daily in cotton fields at the University of Arizona Cotton Research Center,

Phoenix, Arizona, in gossyplure-baited traps were variable. However, average catches of male moths for 3–7 days between boll-sampling periods were strongly correlated with oviposition on cotton bolls, percentages of infested bolls and numbers of larvae per boll. Average weekly numbers of moths emerging from infested cotton were also strongly correlated with the number of males caught. The number of females emerging was strongly correlated with oviposition on cotton bolls. Insecticide applications of carbaryl and fenvalerate reduced catches of male moths of pink bollworm in gossyplure-baited traps compared with catches in traps in untreated fields (Average 56%). However, 13–48 male moths/trap per night were caught in the treated fields after applications. Thus, scheduling treatments on the basis of male moth trap catches, except for the initial treatment, was not feasible. Small field sizes, moth immigration and/or continuing emergence from the infested cotton in the fields may have obscured the impact of the insecticide treatments on adult moth populations.

Riedl *et al*. (1985) developed a procedure to screen field population of codling moth, *Cydia pomonella* (L) (Lepidoptera: Tortricidae), directly for resistance to azinphos-methyl or other toxicants. Male moths were collected in the field with pheromone traps and assayed with azinphos-methyl by topical application to the insects directly on the adhesive coated-trap surface. The LC_{50} of moths trapped during the spring flight was somewhat higher (0.22 µg / moth) than during the summer (0.08 – 0.16 µg / moth). The longevity of moths and their toxicological response was not affected by the polybutene adhesive which was applied as a thin uniform film to the trap surface. Susceptibility to azinphos-methyl increased considerably with age in both sexes and was not correlated with loss in body weight. Haynes *et al*. (1986 a) reported that the effectiveness of combinations of insecticides with pheromones (attracticides) for control of the pink bollworm (PBW) *P. gossypiella* may depend on, among other factors, males freely contacting attracticide source, insecticide-induced mortality, and sublethal interference with the male locating sequence in poisoned males. In flight-tunnel tests, males readily, contacted pheromone sources containing permethrin, fenvalerate, or cypermethrin and suffered mortality. Moreover, after 24 hr survivors were less likely to complete the normal behavioral sequence involved in sex pheromone-mediated mate location. The latter sublethal effects may contribute substantially to the effectiveness of the attracticide technique at the doses of insecticides used in the field. Cypermethrin induced lethal and sublethal effects at a lower concentration than other insecticides tested. Chlordimeform appears to be a poor candidate for attracticide formulations because males avoided contact with this insecticide. Recovery from sublethal effects of cypermethrin occurred after 48 hrs. and represents potential limitations to sublethal modification of behavior for population control.

Haynes *et al.* (1986 b) tested the use of yellow sticky cards to monitor resistance by incorporating various dosages of a selected insecticide into the sticky material on the cards and then seeing if reproducible dose–mortality relationships could be generated from adults flies caught on the cards. The technique was tested with laboratory strains to select the appropriate sticky material, suitable thickness and best time for evaluation of mortality. They found that, the yellow sticky cards are effective when used in monitoring populations of many insect species, and

in monitoring resistance levels to insecticides. Haynes *et al.* (1986 c) described a novel pheromone-baited sticky trap laced with insecticides proved to be a sample and effective means of monitoring insecticide resistance in pink bollworm moth. Adult males from field treated frequently with pyrethroid insecticides showed up to 20–fold resistance to permethrin and up to 14.5-fold resistance to fenvalerate. Information from the resistance-monitoring traps could be used to time the rotation of other chemicals. Miller (1986) reported that a population of tobacco budworm *Heliothis virescens* (Fabricius) from the West Texas cotton fields was found to have a 15-20 fold resistance to pyrethroid insecticides. A new method of measuring resistance in the field, the attracticide method was perfected for adults pink bollworm, *P. gossypiella*. In the attracticide method adult moths collects themselves, sex themselves and dose themselves. The field worker then collects the traps and counts mortality after two days without handling the moths themselves.

Haynes *et al.* (1987) reported that a rapid technique using sex pheromone and insecticide-laced traps was developed for measuring insecticide resistance in pink bollworm moth. This method was developed in the laboratory by allowing males to fly up wind to a sex –pheromone source in a wind tunnel and then trapping them on sticky cards inserted into standard delta traps (delta traps are used for trapping and monitoring Lepidopteran pests or other good-flyer insect pests. They can be used to monitor in orchards, large row crop plantations, small vegetable plots, indoors, etc. Dimensions 12 x 10 x 18 cm. In orange, yellow, or white). Using this technique, populations of adult male *P. gossypiell*a trapped in the field were shown to be more resistant to permethrin and fenvalerate in field frequently treated with pyrethroids than in fields with little or no exposure to these insecticides. The new method eliminates handling of insects that is involved in other methods of assessing toxicity and is compatible with the current practice of monitoring populations with pheromone traps. Schouest and Miller (1988) measured toxicities of fenvalerate and permethrin on a laboratory strain of adult male of pink bollworm by attracticide bioassay. Delta trap was used with a sticky adhesive- coated cards insert containing the insecticide concentration placed in the trap bottom. A rubber septum with 1 mg gossyplure acted as a pheromone source. The caught moths were placed in constant temperature chambers. Data from the attracticide bioassay fit the probit model well and gave a precise estimate of a discriminating dose. Tabashnik *et al*. (1988) monitored resistance in diamondback moth larvae by testing pheromone-attracted males. Three populations were tested for mortality and knockdown responses to fenvalerate, DDT, permethrin and diazinon. Males and females responded similarly. Adult mortality and knockdown were correlated with larval LC_{50} 's across insecticide – population combinations, but adults from one population with DDT- resistant larvae were not resistant to DDT. Brewer and Trumble (1989) developed a technique for monitoring insecticide resistance in field populations of beet armyworm *Spodoptera exigua* (Hubner), and compared with conventional topical bioassay. Inescticides were incorporated into the adhesive of a pheromone trap thereby combining insect collection with insecticide application. This attractant trap technique (ATT) provided stable LC_{50} s with low control mortality when traps were incubated at 21 ºC for 30-36 hrs. after insects were captured. LC_{50} s of a laboratory colony tested with

fenvalerate, permethrin and methomyl were 95.6, 142 and 11.5 µg/g of adhesive, respectively. Slopes of probit regression for males were similar for topical and ATT bioassay indicating a parallel response of the insect to the two methods. Resistance ratios at LC_{50} from (ATT) ranged from 3 to 7.3 for fenvalerate, 1.5 to 2.5 for permethrin and 7.1 to 33 for methomyl.

Campanhola and Plapp (1989) conducted tests to detect reasons for the loss of pyrethroid resistance which occurs in laboratory and field strains of the tobacco budworm in the absence of continuous exposure to pyrethroid insecticides. Attracticide pheromone traps were used in these tests. Larvae of a resistant strain (ICI) maintained in the laboratory developed more slowly than larvae of susceptible laboratory strain. The ICI females were less fertile and produced fewer eggs per individual than susceptible females. In addition, ICI females produced significantly less pheromone and attracted fewer males than did susceptible females. Susceptible males were more attracted to pheromone traps than resistant males. Knight and Hull (1989) investigated the use of sex pheromone traps to monitor the susceptibility of adult male tufted apple bud moths to azinophosmethyl in laboratory experiments and field trials in a number of apple orchards in South Central Pennsylvania and West Virginia. Two techniques were compared: the topical treatment of males caught on the trap ́s sticky surface and the incorporation of the insecticide directly into the adhesive. Compared with the laboratory strain, LD_{50} ̀s populations from apple orchards were 2 to 8 times greater in the topical application bioassays and 6 to 18 times greater in the adhesive incorporation assay. Sanderson *et al*. (1989) used yellow sticky cards with thin layers of insecticide – sticker mixture as a bioassay to determine insecticide resistance in adult *Liriomyza trifolii*, (Burgess) (Diptera: Agromyzidae) , mortality was used to estimate the degree of resistance of a given population to insecticides. Duration of exposure to insecticide before mortality evaluation was standardized at 24 hrs. Control mortality increased with increasing amounts of sticker on the cards and with increasing adult age. Males were slightly more susceptible to permethrin and chlorpyrifos insecticides than females. The sticky card bioassay a simple procedure that is accurate repeatable and usable for field or greenhouse populations. Brewer *et al*. (1990) used a field bioassay that measure adult male susceptibility to document resistance to fenvalerate, permethrin and methomyl in beet armyworm *S. exiqua* . Susceptibility of adult males to these insecticides was monitored in field with technical insecticide mixed into the adhesive of pheromone traps. At the LC_{50}, the highest levels of resistance were typically found at many sites. Geographic and temporal variability in resistance followed this trend: overall variation among regions > variation among sampling dated at the same site within a region ≥ variation among sites in a region within three consecutive days. Knight *et al*. (1990) used sex pheromone traps coated with concentrations of azinophosmethyl impregnated adhesive to test levels of resistance in adult populations of male tufted apple bud moth from apple orchards in seven Eastern States. The results suggest that the level of resistance found within an orchard may be influenced the intensity of fruit production within a region. Level of resistance to azinophosmethyl was positively correlated with current seasonal carbamate use and was not significantly correlated with current use of azinophosmethyl or other organophosphates. Levels of resistance

and fruit injury were both significantly correlated with population densities. Miller (1990) determined the toxicity of carbamate, organophosphorus and pyrethroids insecticides by the attracticide method to field populations of pink bollworm in cotton growing areas of Texas, Arizona, California, Mexico and China. A gradual increase in tolerance to pyrethroid was correlated with high use of these insecticides. A resistance management program for insecticides used in control of pink bollworm is now a possibility and will require considerable cooperation at the grower level. Rummel (1990) reported that during recent years the use of pheromone traps has enabled significant advances to be made in survey, detection and sampling techniques for the boll weevil, pink bollworm, bollworm *Heliothis zea* (Boddie) and tobacco budworm *H. virescens* (F) (Lepidoptera: Noctuidae).

Daly (1992) determined the dose response in different age classes of adults of *H. armigera* (H) by exposing moths to a discriminating dose of fenvalerate using an adult vial test and reported a decline in survival with age so that by 8d after exclusion 60-70% of resistant females and 97% resistant males died at the discriminating dose compared with < 5% of freshly emerged moths. Prabhaker *et al.* (1992) detected resistance in field populations of sweet potato whitefly, efficiently and sensitively using a resistance monitoring technique with yellow sticky cards sprayed with insecticides. Thirty–two populations exhibited various levels of resistance to sulprofos (Resistance ratio [RR] ranging from 19 to 104 and for cypermethrin (RR ranging from 14 to 82) indicating that insecticide resistance is widespread in the Imperial Valley of California. The advantages of this new method for monitoring resistance are discussed in relation to conventional leaf bioassay. Sanderson and Roush (1992) investigated the use of insect trap coated with insecticides mixed with a polybutene adhesive as a resistance monitoring technique for greenhouse whitefly. The technique provided accurate estimates of the mortality response of whiteflies after 24h, with minimal control mortality. The technique was used to estimate diagnostic concentration (LC_{99} of a susceptible strain) and then they were tested against the susceptible and resistant strains. Bush *et al*. (1993) performed a sticky-card bioassay on adult males captured with traps that revealed an 8 fold resistance to parathion in a population of codling moth *C. pomonella* . Parathion resistance in this population was confirmed with a sticky-card bioassay where adult males were exposed to a diagnostic concentration of 120 µg (a.i) parathion per gram adhesive (The estimated LC95 for adult males from susceptible populations). Reduced nonspecific esterase activity detected in adult males captured from the mechanism of codling moth, *C. pomonella* resistance to parathion may be a modified esterase with lower specificity for naphthyl acetate substrates.

Horowitz *et al*. (1993) assayed the efficacy of the insecticide resistance management strategy, a long-term monitoring program in *H. armigera* was undertaken to test the response of this pest to various insecticides. The monitoring technique was based on the exposure of pheromone trap-caught males to groups of insecticides. The diagnostic concentration, established on $LC_{80\text{-}90}$ of a susceptible *H. armigera* field population were 0.57 µg endosulfan, 0.74 µg cypermethrin, 0.38 µg methomyl and 2.0 µg methidathion per scintillation vials. In general, the results indicate that the susceptibility of *H. armigera* to the test compounds was

not appreciably altered from 1987 to 1991, although fluctuations during the season were observed, in most cases, the resistance levels to the test compounds fluctuated in a typical V-shaped curve during the season. The susceptibility of *H. armigera* to the various insecticides observed during the course of this study is consistent with proposition that the insecticide resistance management (IRM) strategy can be correlated with successful management of resistance in this pest. Qureshi *et al.* (1993) used the male moth catches in gossyplure-baited traps to predict larval infestation of pink bollworm, *P. gossypiella* , in cotton fields during 1988 and 1989. The mean moth catches per trap per night were positively correlated with percentage larval infestation. The moth counts in traps and larval infestation in green bolls increased with advance in reproductive stage of the cotton plants. A mean trap catch of 9–12 pink bollworm moths per night was associated with economically damaging infestation. It is, therefore, inferred that insecticidal sprays may be scheduled when 9–12 moths are captured per trap per night.Varela *et al.* (1993) surveyed variation in response to insecticide in codling moth in California, Oregon, Utah and Washington using pheromone traps by two techniques (Topical application and direct incorporation of insecticide into the trap adhesive). LC_{75} from a susceptible population was as a standard dose and 20 apple and pear orchards were monitored for resistance to azinphos-methyl by the topical application. In laboratory tests, female moths were monitored as susceptible as male moths in bioassays with treated adhesive. Anonymous (1995) used an attracticide resistance monitoring technique with a set of key techniques system of integrated management against several damage by the cotton bollworm, *H. armigera* in Xin Xiang, China Demonstration Area, Henan, in the central Yellow River Valley cotton region in 1992 and 1993.Outbreaks of *H. armigera* were effectively controlled and good harvests were obtained using these techniques. These included improved medium term predication techniques, setting up action thresholds, measures to use and conserve natural enemies, systematic monitoring and management of insecticide resistance using pest resistance cotton germplasm and trapping adult noctuids.

Downham *et al.* (1995) described a series of trials examining the feasibility of an attracticide technique for control of *Spodoptera littoralis* (Boisduval). Those trials were assessed by monitoring pheromone trap catches. Similar levels of mating and trap-catch suppression were observed in the two treatments. It was concluded that the mating suppression observed in attracticide plots was due principally to disruption of chemical communication between the sexes, not to male mortality arising from contact with the insecticide sources.Al-Beltagy *et al.* (1996) used pheromone delta sticky traps as an attracticide tolerance monitoring technique in two different locations having two different control strategies for the pink bollworm; one depending on insecticides and the other on pheromone disruption. The field recommended dose as its double and half rates of insecticides dursban and sumicidin were applied to the pheromone delta traps with pheromone sources for the pink bollworm, spiny bollworm. Pheromone tarps as an attracticide tolerance monitoring technique proved to be effective, simple and quick for assaying the moth field strains of Lepidoptera for insecticide resistance. Shekeban (2000) compare the efficiency of the attracticide resistance monitoring technique with different conventional bioassay

techniques and vial technique. Al-Beltagy *et al*. (2001) used attracticide resistance monitoring technique for measuring insecticide resistance in pink bollworm (PBW) male moths, *P. gossypiella* (S). This technique eliminates handling of insect and is compatible with current practice of monitoring populations with pheromone traps. Results showed that, PBW developed resistance to all insecticides tested with R.R. of 19 and 12.7 folds for chlorpyrifos, 14.1 and 34.7 folds for delfose, 14.1 and 38.5 folds for empire and 22.6 and 56 folds for thiodicarb, respectively. This technique used to estimate discriminating concentration for tested insecticides. El-Bassiony (2001) used the attracticide resistance monitoring technique to study pyrethroids resistance under two different pink bollworm control strategies (pheromone disruption technique and insecticides program . El-Armi (2008) used the attracticide resistance mentoring technique to study the development of insecticides resistance against pink bollworm in two different geographical strains.

2. Pheromone traps uses:

2.1. For assaying and monitoring insect pests population density and dynamics all the year round (Al-Beltagy *et al*., 1991). **2.2.** For assaying insect pests geographical distribution throughout its regional and seasonal abundance (Brania and Al-Beltagy, 1996). **2.3.** For insect pests mass trapping technique as a pest control tool (Hamid and Al-Beltagy,1995). **2.4.** For assaying insecticides resistance development in insect pests field strains (Al-Beltagy *et al*., 1996 ; Al-Beltagy *et al.,* 2000 and Al-Beltagy *et al*., 2010). **2.5.** For measuring the successful of a pheromone disruption technique applied against any insect pest (Al-Beltagy *et al*., 2001a and 2001b). **2.6.** For triggering biological and / or chemical control action (Al-Beltagy, 1999). **2.7.** For assaying the efficiency of different insecticides before any insecticide treatment (Al-Beltagy *et al*., 2011).

3. Procedure :

3.1. Attracticide resistance monitoring technique (ARMT) :

A novel resistance monitoring method was created and perfected for pink bollworm by Miller (1986) and Haynes *et al.* (1986 a and 1987) and was modified by Al-Beltagy *et al.*(1996) and Shekeban (2000) under the Egyptian conditions. This method employs pheromone baited delta traps (Figure 1) that are usually used for assessment of populations of male moths. The conventional yellow paper delta traps supplied by the Ministry of Agriculture in Egypt are used for the population assessment in different cotton fields all over the country.

3.2. Laboratory bioassays:

For ARMT laboratory bioassays, serial concentrations of each tested formulated pyrethroid and organophosphorous are diluted in acetone and mixed with sticky material. Condensed petroleum glue, as a trapped sticker, produced by Agrycins Corp., England, and supplied by Ministry of Agriculture, are used. Two grams of the sticky adhesive and different insecticide concentrations for pyrethroids ranged from 0.05 to 20 ppm and for OP's ranged from 1 to 240 ppm. Each concentration for each insecticide is replicated three times on separate sheet card. Control without insecticide is included. Male moths are transferred from the rearing facility to the bioassay laboratory. Ten male moths are captured and sticked to the yellow sticky card which is treated before with the insecticide concentration as mentioned. The insert cards are kept under constant conditions of 21 ± 3°C and 95 ± 5% R.H. Adult mortalities are recorded at 6 and 12 h-periods. Mortality is subjected

for probit analysis to estimate LC_{50}, LC_{95} and slope. Resistance ratios (R.R.) at LC_{50} (or LC_{95}) are calculated by dividing each value for field strain, of each species, by corresponding value of laboratory reference strain.

3.3. Preparation of insecticide-pheromone-baited sticky trap:

For preparation of pheromone-baited sticky trap laced with an insecticide the following technique is used: serial concentrations of each tested pyrethroid and organophosphorous formulations are diluted in acetone and mixed with sticky material. Condensed petroleum glue, as a trapped sticker, produced by Agrycins Corp., England, and supplied by Ministry of Agriculture, is used. Two grams of the sticky adhesive and an insecticide concentration are scarped on the trap insect sheet cards (Inserts) using putty knife starting with the lowest concentration and ending to the highest one, every time the putty knife is washed with kerosene and dried. Six diluted concentrations from each insecticide are used, with relative folds of the field recommended rate (F). 0.125 F, 0.25 F, 0.5 F, 1 F, 1.5 F and 2 F. These inserts (cards) are secured with paper for attracticide resistance monitoring technique (ARMT) field bioassays, the traps are baited with an appropriate commercially available pheromone lures (Figure 2) supplied by the Ministry of Agriculture in Egypt, for trapping techniques.

3.4. Field experiments:

The field experiments are carried out twice during the growing seasons: 1) at the early-season when plant buds emerged, and 2) at the late-season when the fruits are up to the maturity as following steps: - Traps are hung on stakes higher 20-30 cm above the canopy of the plants and distributed by a rate of one trap / five feddans. - The dosed insert cards and control cards are placed into the delta traps with a pheromone source, randomly. All cards are inserted before sunset.

- Traps are left overnight and collect quickly in the following morning. - All insert cards are collected at dawn with trapped adult male moths (Figure 3) and checked for mortality %, this termed the zero time mortality %, then placed each insert card into the holding container (Figure 4) . - The cards are collected at dawn to avoid the heat of the day, the trapped moths will not be well if they were allowed to warm up in the morning sun. - Cards are kept away of the sun in the shadow until they delivered from the field to their storage sites.- It is very important to store the insert cards in the holding containers in a constant temperature 21 ± 3 ºC for 12 hours. After 6 hr and 12 hr cards are removed from container and mortality are recorded. - The adults are scored as alive if any movement is recorded (i.e., leg, abdomen, wing even the antenna). The insects are recorded as died if they do not move at all. - Each dose and control are replicated three times.- When all doses and cards are accounted and all insects are checked for mortality, these data together with the mortality of controls for each insecticide are analyzed by probit analysis computer program (Finney, 1971) to determine slope, LC_{50} and LC_{95} values. Resistance ratios (RR) at LC_{50} or LC_{95} are calculated by dividing LC_{50} for field strain, of each species, by LC_{50} of laboratory (Reference) strain. Abbott's formula (Abbott, 1925) is used to correct mortality according to mortality control traps.

4. Biochemical confirmations :

Biochemical studies (Enzymes and /or proteins) are conducted for the confirmation of insecticides resistance development in insect pest field strains data that are recorded by the pheromone traps technique, throughout enzymes activity determinations and total protein electrophoretic assays (Al-Beltagy,

1993, Albeltagy *et al*., 2001a and Albeltagy *et al*., 2001 b).

5. The recommendation:

Many advantages have been indicated for the use of the attracticide resistance monitoring technique (ARMT), some of these advantages – as a tool for indicating the efficacy of different insecticides are :- **5.1.** It is easy, whereas no need to complicate equipment or high technology tools, but only use some delta pheromone traps. **5.2.** It is speed, whereas it is applied by sunset and get data by next morning sunrise, just after about 12 hours of applications. **5.3.** It is effective, whereas it is applied in a very short period (As mentioned above), so there is no interference of any changeable environmental conditions . **5.4.** It is applied directly against the field insect population strains, whereas no need for the mass rearing techniques and facilities.**5.5.** It is very accurate – for the above mentioned advantages – compared to the conventional insecticide bioassay techniques which depend on the application against insect colonies that are got from rearing facilities for a few generations which cause that these strains may be very closed to the susceptible strains.

Figure (1): Yellow paper delta pheromone traps over cotton plants.

Figure (2) : Rubber septum pheromone sources.

Figure (3): Adult moths sticked to sticky sheet.

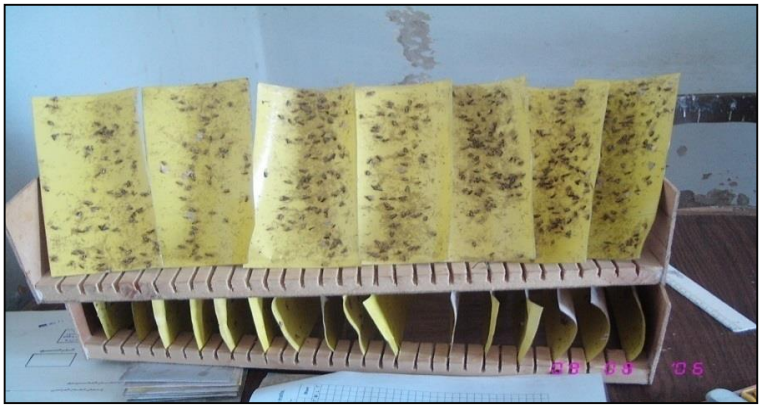

Figure (4): Adult moths holding container.

References

Abbott, W.S. (1925): A method of computing the effectiveness of an insecticide. J. Econ. Entomol., 18 : 265-267.

Al-Beltagy, A.M. ; El-Mourshedy, and Mourad, A.K. (1996): Pheromone traps for monitoring insecticide tolerance in three different lepidopterous insects: an effective tool for insecticide resistance management. Egypt. J. Agric. Res., 74 (1): 29-37.

Al-Beltagy, A. M.; Radwan, H. S. ; El-Bermawy, Z. A. ; Nassar, M. E. ; Yousef, A. G. and Shekeban, M. M. (2001): Attracticide resistance monitoring technique for assaying insecticide resistance in pink bollworm male moths, *Pectinophora gossypiella* (S) field populations. Egypt. J. Agric. Res., 79 (3): 949-963.

Al-Beltagy, A.M. ; Shawir, M. S. ; Osman, K.A. ; Abo El-Saad, M.M. and Mourad, M. A. (1993): Biochemical technique for measuring susceptibility of pink bollworm, *Pectinophora gossypiella* to certain insecticides. Alex. J. Agric. Res., 38 (3): 393-411.

Al-Beltagy, A.M.; Radwan, H.S. ; Abo El-Ghar, G.E. ; Ammar, I.M. ; Eisa, A.A. ; Shekiban, M.M. and El Bassiony, M.M. (2001a): Biochemical impact of pyrethroid resistance management in bollworms field populations with Gossyplure disruption technique .1- in

relation to total protein electrophoretic analysis. J. Adv. Agric. Res. , 6(4) : 1029 –1041 .

Al-Beltagy, A.M.; Radwan, H.S. ; Abo El-Ghar, G.E. ; Ammar, I.M. ; Eisa, A.A. ; Shekiban, M.M. and El Bassiony, M.M. (2001b) . Biochemical impact of pyrethroid resistance management in bollworms field populations with Gossyplure disruption technique .2- in relation to AchE and total esterases activity determination. J. Adv. Agric. Res. , 6(4) : 1043 – 1055 .

Al-Beltagy, A.M.; Shaker, N.; Taman, F.A. and El-Khishen, S.A. (1991): The influence of pheromone traps on the infestation percentages and population density of bollworms in Alexandria and Al-Behera Governorates. Proc. of the 4th Arab Congress of Plant Prot., Cairo, 1-5 Dec., 7 (3): 477-487.

Al-Beltagy, A.M.; Shawir, M.S.; Othman, E.A.; Abo-Elsaad, M.M. and Mourad, M.A. (1993): Biochemical techniques for measuring susceptibility of pink bollworm, Pectinophora gossypiella to certain insecticides., Alex. J. Agric. Res., 38 (3): 393-411.

Al-Beltagy, A.M.; Shekeban, M. M.K. ; Abo El-Amayem, M.M.; Kassem, S. M. I. ; Mancee, A.H. and EL-Arami, S.A. (2010 : Monitoring for Pyrethroid resistance in pink bollworm *Pectinophora gossypiella* (Saunders) field strains . Pest Management Newsletter , 19(2) : 46-52 .

Al-Beltagy, A.M; Abo El-Amayem, M.M.; Kassem, S. M. I.; Mancee, A.H.; Shekeban, M. M.K. and EL-Arami, S.A. (2011): Assaying insecticidal efficacy of some formulated organophosphorus insecticides against pink bollworm male moth field strains using the attracticide efficacy monitoring technique. Resistant Pest Management Newsletter, 21 (2): 7-13.

Al-Beltagy; A.M (1999): Plant and insect triggers for sequential use of gossyplure and insecticides for pink bollworm control actions . Egypt J. Agric. Res., 77(4) : 1633 – 1643 .

Anonymous (1995): A study of the key technique system of integrated management against severe damage by the cotton bollworm. CAB Abstracts; record 9 of 19.

Brania , H. A. A. and Al-Beltagy, A.M. (1996): Regional and seasonal variations of pink bollworm trap catches and boll infestation relationships. Alex. J. Agric. Res., 41 (3): 77-84.

Brewer, M.J. ; Trumble, J.T.; Rodrigues, B.A. and Chaney, W.E. (1990): Beet armyworm adult and larval susceptibility to three insecticides in managed habitats and relationship to laboratory selection for resistance. J. Econ. Entomol., 83(6):2136-2146.

Brewer, M.J. and Trumble, J.T. (1989): Field monitoring for insecticide resistance in beet armyworm. J. Econ. Entomol., 82(6): 1525-1526.

Bush, M.R.; Abd El-Aal, Y.A.I. and Rock, G. C. (1993): Parathion resistance and esterase activity in codling moth (Lepidoptera: Totricidae) from North Carolina J. Econ. Entomol., 86(3):660-666.

Campanhola, C. and Plapp, Jr F.W. (1989): Pyrethroid resistance in the tobacco budworm

(Lebidoptera: Noctuidae) insecticide bioassay and field monitoring. J. Econ. Entomol., 82(1):22 -28.

Daly, J.C. (1992): Decline in survival with age of resistance adult *Helicoverpa armigera* (Lepidoptera : Noctuidae) at a discriminating dose of the pyrethroid fenvalerate**.** J. Econ. Entomol., 83(3):705 -709.

Downham, M.C.A. ; Mc Veigh, L.J. and Moawad, G.M. (1995): Field investigation of an attracticide control technique using the sex pheromone of the Egyptian cotton leafworm *Spodoptera littoralis* (Lepidoptera : Noctuidae). Bull. Entomol. Res., 85: 463-472.

El-Armi, S. A. A. (2008): Insecticides efficacy of some insecticide formulations under certain environmental conditions. Ph. D. Thesis, Pesticides Chemisry, Fac. Of Agric., Alexandria University.

El-Bassiony M. M. (2001): Studies on the management of pyrethroid resistance in bollworm populations in cotton fields. Ph. D.Thesis, Pesticides Department, Menoufiya University.

Finney, D. J. (1971): Probit analysis. A Statistical Treatment of the Sigmoid Response Cure. 7th Ed., Cambridge Univ. Press, England.

Georghiou, G.P. (1983): Management of resistance in arthropods pp. 769-792. In G.P. Georghiou and T. Satto (eds), pest resistance to pesticides. Plenum, New York.

Hamid, A.M. and Al-Beltagy, A.M. (1995): Mass-trapping technique for pink bollworm control. Proc. of 1st Int. Conf. of Pest Control, Mansoura, Egypt, 3-5 Sept: 189-195.

Haynes, K.F. ; Li, W.G. and Baker , T.C. (1986 a): Control of pink bollworm moth (Lepidoptera: Gelechiidae) with insecticides and pheromones (Attracticide): lethal and sublethal effects. J. Econ. Entomol., 79 (2): 1466-1471.

Haynes, K.F. ; Miller, T.A. ; Staten, R.T. ; Li, W.G. and Baker, T.C. (1987): Pheromone traps for monitoring insecticide resistance in the pink bollworm moth (Lepidoptera : Gelechiidae) : New tool for resistance management. Environ.Entomol., 16:84-89.

Haynes, K.F. ; Miller, T.A. ; Staten, R.T. ; Li, W.G. and Baker, T.C. (1986 c): Monitoring insecticide resistance with insect pheromone. Experientia., 42:1293-1295.

Haynes, K.F.; Parrella , M.P.; Tumble, J.T. and Miller, T.A. (1986 b): Monitoring insecticide resistance with yellow sticky cards. California Agric., 11-12.

Henneberry, T.J. and Clayton, T. E. (1982): Pink bollworm of cotton (*Pectinophora gossypiella* (Saunders)): male moth catches in gossyplure-baited traps and relationships to oviposition, boll infestation and moth emergence. Crop Protection., 1(4):497-504.

Horowitz, A.R. ; Seligman, I. M. ; Forere, G. ; Bar, D. and Ishaaya, I. (1993): Preventive insecticide resistance strategy in (*Helicoverpa) Heliothis armigera* (Lepidoptera: Noctuidae) in Israel cotton. J. Econ. Entomol., 86 (2) : 205-212.

Knight, A. L ; Hull, L.A. ; Rajotte, E. ; Hogmire, H. ; Horton, D. ;

Polk, D. ; Walgenbach, J.; Weires, R. and Wnalon, J. (1990): Monitoring azinphos-methyl resistance in adult male *Platynota idaeusalis.* (Lepidoptera : Tortricidae) in apple from Georgia to New York. J. Econ. Entomol., 83(2):329-334.

Knight, A. L. and Hull, L. A. (1989): Sex pheromone traps to monitor azinphosmethyl resistance in tufted apple bud moth (Lepidoptera: Tortricidae). J. Econ. Entomol., 82(4): 1019-1026 .

Miller, T.A. (1986): Status of resistance in the cotton insect complex. Beltwide Cotton Production Research Conf., 162-165.

Miller, T.A. (1990): Resistance monitoring of pink bollworm California Department of Food and Agriculture (CDFA), pink bollworm final report , 146-147.

Mink, J.S. and Boethel, D.J. (1992): Development of a diagnostic technique for monitoring permethrin resistance in soybean looper larvae (Lepidoptera : Noctuidae). J.Econ. Entomol., 85(4):10565-10620.

Pasquier, D. and Charmillot, P.J. (2003): Effectiveness of twelve insecticides applied topically to diapausing larvae of the codling moth, *Cydia pomonella* L. Pest. Manag. Sci., 60 (3):305–308.

Pimentel, D. (2005): Environmental and economic costs of the application of pesticides primarily in the United States. Environ. Develop. and Sustain., 7 (2): 229-252.

Prabhaker, N. ; Toscano, N.C. ; Perring, T.M. ; Nuessley, G. ; Kido, K. and Youngman, R.R. (1992): Resistance monitoring of the sweet potato whitefly in the imperial valley of California. J. Econ. Entomol., 85 (4):1063-1068.

Qureshi, Z. A. ; Ahmad, N. and Hussain, T. (1993): Pheromone trap catches as a means of predicting damage by pink bollworm larvae in cotton. Crop. Protection, 12(8): 597-600.

Riedl, H. ; Seaman, A. and Henrie, F. (1985): Monitoring susceptibility to azinphos-methyl in field population of the codling moth (Lepidoptera: Tortricidae) with pheromone traps. J. Econ. Entomol., 78 (1) : 692-699.

Roush, R. T. and McKenzie, J. A. (1987): Ecological genetics of insecticide and acaricide resistance. Ann.Rev. Entomol., 32: 361-380.

Roush, R.T. and Miller, G.L. (1986): Considerations for design of insecticide. J. Econ. Entomol., 87 (2): 184-192.

Rummel, D.R. (1990): **Pheromone in pest detection and pest management, CDFA, pink bollworm final report., 145-146.**

Sanderson, J.P. and Roush, R.T. (1992): Monitoring insecticide resistance in green house whitefly with yellow sticky cards. J. Econ. Entomol., 85(3):634-642.

Sanderson, J.P. ; M.P. Parrella and J.T. Trumble (1989). Monitoring insecticide resistance in *Liriomyza trifolii* (Diptera : Agromyzidae) with yellow sticky cards. J. Econ. Entomol., 82(4):1011-1018.

Schouest, L.P. Jr. and Miller, T.A. (1988): Factors influencing pyrethroid toxicity in pink bollworm (Lepidoptera :

Gelechiidae) : Implications for resistance management. J. Econ. Entomol., 81(2):431-436.

Shekeban, M. M. K. (2000): Studies on insecticide resistance in certain major cotton insects Department of Pesticides, Faculty of Agriculture, Menoufiya University.

Tabashnik, B.E. ; Rethwisch, M.D. and Johnson, M.W. (1988): Variation in adults mortality and knockdown caused by insecticides among populations of diamondback moth (Lepidoptera : Plutellidae). J.Econ. Entomol., 81(2):437-441.

Tabashnik, B.E. and Cushing, N.L. (1987): Leaf residues topical bioassay for assessing insecticide resistance in the diamondback moth. FAO plant Prot. Bull., 35:11-14.

Tamhankar, A.J. ; Gahukar, R.T. and Rajendran, T.P. (2000): Pheromones in the management of major lepidopterous and coleopterous pests of cotton. Integr. Pest. Manag. Rev., 5 (1) :11-23.

Varela, L.G.; Wekter, S.C.; Jones, V.P.; Brunner, J.F. and Riedl, H. (1993).: Monitoring and characterization of insecticide resistance in codling moth (Lepidoptera : Torticidae) in four Western States. J. Econ. Entomol., 86 (1): 1-10.