

## Mortality Comparison Among Spinosad-, Actara-, Malathion-, and Methomyl-Containing Baits Against Peach Fruit Fly, *Bactrocera zonata* Saunders (Diptera: Tephritidae) Under Laboratory Conditions

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**Abstract:** Peach Fruit Fly (PFF), *Bactrocera zonata* (Diptera: Tephritidae) is a newly recorded species of fruit flies in Egypt last decade. The toxicity of spinosad, actara, malathion and methomyl baits was assessed against male and female adults of PFF under laboratory conditions. In a laboratory feeding assay, the carbamate insecticide, methomyl affected flies more than the neonicotinoid, actara, the biorational insecticide, spinosad, and the OP-insecticide, malathion. The LC50 values of Methomyl against the adult males of PFF were 0.169 and 0.041 ppm 24 and 48 hrs posttreatment, respectively. Among the female adults, the LC50 values were 0.351 and 0.076 ppm 24 and 48 hrs posttreatment, respectively. While the LC50 values of Actara against the adult males of *B. zonata* were 9.166 and 6.383 ppm 24 and 48 hrs posttreatment, respectively. Among female adults, the LC50 values were 11.154 and 7.024 ppm 24 and 48 hrs posttreatment, respectively. Present results showed that the LC50 values of spinosad for males were 20.05 and 16.00 ppm 24 and 48 hrs posttreatment, respectively. The LC50 values for female flies were 26.96 and 19.11 ppm 24 and 48 hrs posttreatment, respectively. The LC50 values of Malathion against male and female adults of *B. zonata* were 206 and 75.3 ppm for adult males, and 345.1 and 83.8 ppm for adult females, 24 and 48 hrs posttreatment, respectively. It is observed also that LC50 values of all tested compounds were higher in the case of female than in male adults. Spinosad was more potent than the fruit fly-recommended insecticide, malathion. Although having the most rapid onset of activity in laboratory tests, up to 23% of flies remained alive after exposure to malathion bait. In contrast, <12% of flies survived 24-h exposure to spinosad or actara baits. Given its favorable safety profile, a high degree of initial activity comparable with malathion and lack of cross-resistance to other chemistries, spinosad seems to be promising as a bait spray additive and replacement for Malathion for peach fruit fly, *B. zonata* control programs to help control or delay the emergence of resistant populations.

**Key words:** Spinosad, Actara, Malathion, Methomyl, Tephritidae, Peach Fruit Fly, *Bactrocera zonata*

### INTRODUCTION

Fruit flies (Diptera; Tephritidae) rank among the world the most serious pests of horticultural crops. In addition to, causing direct losses in the yield and marketability, they pose as significant threats to quarantine security and thus to international trade in fruits and fresh vegetables world-wide<sup>[19]</sup>. Four hundred species belonging to the genus *Bactrocera* are widely distributed in tropical Asia, South Pacific and Australia regions, but very few species of such genus were recorded in Africa<sup>[8]</sup>.

Peach fruit fly (PFF), *Bactrocera zonata* (Saunders), is one of the most serious polyphagous insect pests<sup>[14]</sup>. It attacks a large host range of fruit and vegetables hosts; such as mango, peach, fig, guava, citrus, tomato and apple<sup>[20,40]</sup>.

PFF is a newly recorded species of fruit flies in Egypt last decade. The fly became widespread over different locations in Egypt, such as Alexandria<sup>[10]</sup>, Kalubeia<sup>[6]</sup>, and El-Beheira<sup>[6]</sup> Governorates. The presence of certain fruit flies in a country can pose a significant barrier to trade in fresh fruits and vegetables. Since this species is gradually increases its host range, their significant damage to Egyptian agriculture is increasing year after year. Because newly fields are adding to the orchards, overcropping operations, also the difficulties to choose suitable methods of control in view of the awareness of problems associated with the use of insecticides as cover spray. However, little information about using bait application technique (BAT) for controlling of PFF, so far, is available.

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The application of insecticides is an essential component for eradication or management of fruit fly pests. Impact on nontarget organisms and public rejection of area wide pesticide applications have been major concerns in managing these programs. Bait stations have been proposed as alternative treatments in areas where broadcast insecticides are not acceptable<sup>[21]</sup>.

Current PFF control in Egypt is based primarily on applications of organophosphate insecticides, especially malathion, mixed with protein baits. The intensity of insecticide treatments with malathion against fruit flies has resulted in the development of resistant populations<sup>[29]</sup>. Moreover, the use of malathion is controversial because of human health concerns<sup>[13,23]</sup> and the harmful effects it has on beneficial insects<sup>[5,39]</sup>. In recent years, emphasis has been placed on implementing safer environmental measures to control the fruit flies. To this end, we are testing insecticides that are more environmentally friendly than malathion. One of the insecticides considered is spinosad<sup>[1,34]</sup>. Compared to malathion, this insecticide has a better environmental profile and is less toxic to natural enemies<sup>[39,41,35]</sup>. At present, a spinosad bait treatment, containing spinosad and a mix of sugars, water and attractants, is successfully being used to control different tephritid pests worldwide<sup>[3]</sup>.

The present study aims to compare, in the laboratory, the efficacy of some organophosphate and carbamate insecticides as well as some bio-insecticides at various concentrations on male and female adults, away to application BAT to control PFF in the field as partial spray to avoid the problems resulted from the application of cover spray procedure.

## MATERIALS AND METHODS

**Laboratory Assay of Some Selected Insecticides Against Adults of PFF:** The experiment was planned to evaluate the toxicity of various insecticides against male and female adults of PFF, *B. zonata*.

**Laboratory Rearing Technique:** The initial culture of PFF, *B. zonata*, has been obtained from infested mango fruits which were collected from a farm at Kom-Hamada district (60 Km southward of Damanhour City) in August 2004. The insect was reared in the laboratory according to the rearing method described by El-Aw *et al.*<sup>[9]</sup>. According to such method, the infested fruits were kept under the laboratory conditions ( $26.0 \pm 2.0^\circ\text{C}$  and  $70.0 \pm 5.0\%$  R.H.). Plastic jars were furnished by sterilized sand and the infested fruits were placed inside the jars until pupation. Pupae were collected daily and transferred to adult rearing cages (30 X 30 X 30 cm). The different sides of the adult

cages were coated with wire screen except one side which has a sleeve opening (for daily examination) and the cage floor was made of wooden sheet.

**Mass Rearing Technique:** Pupae of PFF were kept under the above-mentioned laboratory conditions. The newly emerged flies were provided with adult food consists of sugar mixed with hydrolyzate protein (3:1w/w) and wet cotton as a source water. The adults of both sexes were mated in the cages. Cups of Guava juice covered with muslin were placed in the cages as an oviposition site. The deposited eggs were collected every 24 hours and washed with tape water. The collected eggs were placed on an artificial diet contains molasses (125 gm), wheat bran (1500 gm), tap water (500 ml), sodium benzoate (6 gm), citric acid (4 gm) and brewer yeast (75 gm) as described by El-Aw *et al.*<sup>[9]</sup>. The diet was kept in plastic jars and stored in refrigerator until use.

The eggs were placed on the surface of the diet inside plastic cups (9 cm diameter and 5 cm height). These cups were covered for the first three days with pieces of thick cloth lid to safe the moisture in the cups for egg hatching. During the larval growth the thick cloth was replaced with muslin fabrics until larval full-grown then the muslin lids were removed and the cups which containing the grown larvae were put inside large plastic jars furnished with sand. The matured larvae were jumped out and pupated in the sand. After complete pupation the sand was sieved and the pupae collected, then placed in a petri dish and transferred to the rearing cages to start a new generation. The newly emerged flies were used for evaluating the toxicity of various insecticides against male and female adults of PFF, *B. zonata*.

**The Insecticides:** The tested insecticides belong to different classes; Organophosphate (Malathion 57% EC), Neonicotinoid (Actara, Thiamethoxam 25 % WS), Carbamate (Methomyl 90 % WP), and biorational-insecticide (Spinosad 24% SC) (Produced by Dow AgroSciences Co.) and each of the tested insecticides were mixed with the attractive feeding material (buminal).

**The Procedure:** Ten male and female flies of PFF (6-days-old) were confined separately in plastic cup coated with muslin, without food, for 12 hrs and provided with water. Pieces of fiberboard (1 x 1 x 0.25 cm) were immersed for 10 seconds in a series of different concentrations of the tested insecticide. After 5 minutes, the fiberboards were placed on the lids of the cups. Three replicates were used for each concentration, either for male or female flies.

The cups were examined after 24 and 48 hrs and the died flies were counted and recorded. The average percentage of adult mortality for each concentration was calculated and plotted against each test concentration on a logarithmic probit paper. The concentration required to kill 50 % of tested adults (LC<sub>50</sub>) was calculated according to Finney<sup>[12]</sup>. The relative efficiency of the tested insecticides was determined according to<sup>[37]</sup>.

## RESULTS AND DISCUSSION

**Efficiency of Some Selected Insecticides for Control of Adults of PFF:** Mortality percentages (M %) of male and female adults of PFF exposed to a series of concentrations of some selected compounds belong to different insecticide classes are presented in Tables (1-4). The toxicity of Methomyl, Actara, Spinosad and Malathion, individually mixed with buminal, was evaluated in a laboratory bioassay.

Numbers of dead flies were counted and recorded and mortality percentages of adults of both sexes of PFF were calculated 24 and 48 hrs posttreatment with the above-mentioned tested insecticides. Obtained data demonstrated that the adult susceptibility to the different tested insecticides was found to be different based on the calculated LC<sub>50</sub> values.

Table (1) shows the LC<sub>50</sub> value of the Carbamate insecticide, Methomyl, against the adult males of PFF was 0.169 ppm 24 hrs posttreatment, which decreased to 0.041 ppm 48 hrs posttreatment. Among the female adults, the LC<sub>50</sub> values were 0.351 and 0.077 ppm 24 and 48 hrs posttreatment, respectively. It is clear that adult mortalities were increased not only by Methomyl concentrations but also by increasing the days posttreatment.

Data in Table (2) show that the LC<sub>50</sub> value of the neonicotinoid insecticide, Actara, against the adult males of *B. zonata* was 9.166 ppm 24 hrs post treatment which decreased to 6.604 ppm at 48 hrs posttreatment. Among female adults of *B. zonata*, the LC<sub>50</sub> values were 11.154 and 7.021 ppm 24 and 48 hrs posttreatment, respectively. It is clear that the LC<sub>50</sub> values were higher at 24 hrs than at 48 hrs posttreatment with the tested insecticide, Actara, either for male or female adults. It is observed also that such LC<sub>50</sub> values were higher in the case of female than in male adults.

On the other hand, in the case of application of the biorational insecticide, Spinosad, data in Table (3) show that the LC<sub>50</sub> values for male flies were 20.05 and 15.99 ppm 24 and 48 hrs posttreatment, respectively, with Spinosad. Results showed also, that the LC<sub>50</sub> values for female flies were 27.00 and 19.11 ppm 24 and 48 hrs posttreatment, respectively.

Table (4) shows also the LC<sub>50</sub> values of the OP insecticide, Malathion, against the adult males and females of *B. zonata*. Such LC<sub>50</sub> values 24 and 48 hrs posttreatment were 205.95 and 75.33 ppm for adult males, and 345.1 and 83.85 for adult females, respectively. Such values indicated that the LC<sub>50</sub> values recorded after 24 hrs posttreatment were about 3-4 times higher than those recorded at 48 hrs.

Data in Table (5) show the LC<sub>50</sub> values and toxicity index for the tested compounds against the male and female adults of *B. zonata*. The toxicity index values show superior efficiency (100%) of Methomyl at LC<sub>50</sub>s for male and female adults of *B. zonata* at 24 and 48 hrs posttreatment. Toxicity index values were 1.84 and 0.642 % at 24 and 48 hrs posttreatment for treated males, respectively. Such values were increased to 3.147 and 1.113 % for female adults treated with the insecticide, Actara, at 24 and 48 hrs posttreatment, respectively. Thus, the efficiency ratios of Actara female LC<sub>50</sub> / male LC<sub>50</sub> were 1.216- and 1.100-folds at 24 and 48 hrs posttreatment, respectively. Whereas toxicity index values for Spinosad were 0.84 and 0.285 % at 24 and 48 hrs posttreatment for treated males, respectively, and then increased to 1.302 and 0.418 % for treated females at 24 and 48 hrs, respectively. The efficiency ratios of Spinosad female LC<sub>50</sub>/male LC<sub>50</sub> were 1.35- and 1.29-folds at 24 and 48 hrs posttreatment, respectively.

The toxicity index values for Malathion were 0.082 and 0.054 % for male adults and then increased to 0.102 and 0.093 % for female adults of *B. zonata* at 24 and 48 hrs posttreatment, respectively. Accordingly, the efficiency ratios of Malathion female LC<sub>50</sub> / male LC<sub>50</sub> were 1.67- and 1.11-folds 24 and 48 hrs posttreatment, respectively. This indicates that the LC<sub>50</sub> values of 24 hrs posttreatment are about 3.4-times higher than those of 48 hrs. It means that the adult male flies were more susceptible than adult female flies of *B. zonata*. However, present findings confirm the results of Stark *et al.*<sup>[25]</sup> that adult males of *C. capitata* were significantly more susceptible to Spinosad than females. The authors found that male and female flies of *B. cucurbitae* and *B. dorsalis* were equally susceptible to Spinosad insecticide.

It is observed from Tables (1-4) that the slopes of the lcp-lines of treated adults with each of the tested insecticides differed between both *B. zonata* sexes and between 24 hrs to 48 hrs posttreatment within each sex. Such slope values were also differed from one tested insecticide to another. However, except in the case of Actara, the slope values of the lcp-lines were generally higher at 24 than at 48 hrs. The slope values, in most cases, were increased in the following order: Methomyl, Malathion, Spinosad and Actara. Present results indicate that the heterogeneity of the treated

**Table 1:** Mortality percentages, LC<sub>50</sub> and slope values for male and female adults of *B. zonata* after feeding on different concentrations of Methomyl-bait under laboratory conditions.

Insecticide	Concentration (ppm)	Mortality %			
		Males		Females	
		24 *	48	24	48
Methomyl	10.0	99.90	100	93	100
	5.0	80.90	89.00	77.83	89.60
	0.5	66.00	79.00	55.33	78.00
	0.1	45.33	60.00	33.00	55.00
	0.01	13.66	33.16	11.33	20.00
	LC <sub>50</sub>	0.169	0.041	0.351	0.077
	Upper limit	0.561	0.069	0.502	0.120
	Lower limit	0.030	0.021	0.242	0.048
	Slope	0.855	0.640	0.814	0.780

Time posttreatment (hrs)

**Table 2:** Mortality percentages, LC<sub>50</sub> and slope values for male and female adults of *B. zonata* after feeding on different concentrations of Actara-bait under laboratory conditions.

Insecticide	Concentration (ppm)	Mortality %			
		Males		Females	
		24 *	48	24	48
Actara	40	88.66	99.66	77.33	90.00
	20	66.33	76.33	66.00	76.00
	10	55.00	65.00	44.00	55.00
	5	33.00	44.00	33.00	44.00
	2.5	16.91	21.17	18.57	22.86
	LC <sub>50</sub>	9.166	6.604	11.154	7.021
	Upper limit	11.008	7.797	13.68	8.363
	Lower limit	7.317	5.502	9.689	5.787
	Slope	1.696	1.742	1.376	1.584

Time posttreatment (hrs)

adults of *B. zonata* towards the tested insecticides is increased in the opposite order; Actara, Spinosad, Malathion and Methomyl. This may also indicate heterogeneity in the susceptibility levels of their biochemical systems toward the different tested insecticides. Obtained data demonstrated also that the LC<sub>50</sub> values for male adults were less than those of female adults either at 24 or at 48 hrs posttreatment. It is concluded from the present results that female adults of *B. zonata* are less sensitive to Methomyl, Actara, Spinosad and Malathion insecticide than those of male ones. Generally, it is indicated that Methomyl was the most effective insecticide against male and female adults of *B. zonata* followed by Actara, Spinosad and Malathion.

Present work tested the use of food attractive-insecticide-bait (bait application technique, BAT) for controlling *B. zonata*. BAT is a widely used technique for controlling fruit flies<sup>[11]</sup>. According to Manra-khan and Price<sup>[22]</sup>, such BAT included the use of protein-insecticide-bait spray. However, Malathion is usual choice of insecticide for fruit flies control and this is usually combined with protein hydrolysate to form a bait spray<sup>[31]</sup>. Several authors used Malathion insecticide bait against fruit flies such as *B. zonata*<sup>[15]</sup>, *C. capitata*<sup>[30]</sup>, *B. zonata*, *B. cucurbitae* and *Carpomyia vesuviana*<sup>[36]</sup>. However, our results indicated that Malathion was the least effective tested compound among the tested insecticides based bait against *B. zonata*. It is clear that female flies are more tolerant to

**Table 3:** Mortality percentages, LC<sub>50</sub> and slope values for male and female adults of *B. zonata* after feeding on different concentrations of Spinosad-bait under laboratory conditions.

Insecticide	Concentration (ppm)	Mortality %			
		Males		Females	
		24 *	48	24	48
Spinosad	120	88.66	92.00	88.66	90.00
	60	77.33	81.00	66.33	76.00
	30	66.00	70.00	55.00	65.00
	20	44.00	50.00	44.00	54.00
	10	33.00	40.00	22.00	30.00
	LC <sub>50</sub>	20.054	15.99	27.00	19.11
	Upper limit	23.97	17.25	31.72	22.87
	Lower limit	16.154	12.39	22.70	15.32
	Slope	1.583	1.560	1.690	1.590

Time posttreatment (hrs)

**Table 4:** Mortality percentages, LC<sub>50</sub> and slope values for male and female adults of *B. zonata* after feeding on different concentrations of Malathion-bait under laboratory conditions.

Insecticide	Concentration (ppm)	Mortality %			
		Males		Females	
		24 *	48	24	48
Malathion	1000	80.50	90.00	78.33	92.33
	500	63.00	76.00	57.66	80.66
	250	54.00	60.00	41.42	71.33
	100	40.00	50.00	20.66	55.33
	50	22.00	40.00	10.00	38.83
	LC <sub>50</sub>	205.95	75.33	345.06	83.85
	Upper limit	269.11	104.53	418.67	108.34
	Lower limit	156.6	48.129	289.04	59.84
	Slope	1.150	1.039	1.590	1.230

Time posttreatment (hrs)

**Table 5:** LC<sub>50</sub> and toxicity index values for treated PFF, *B. zonata*, adults with some different insecticide-baits under laboratory conditions.

Compound	LC <sub>50</sub>				Toxicity Index at LC <sub>50</sub>			
	Male		Female		Male		Female	
	24 *	48	24	48	24	48	24	48
Methomyl	0.169	0.041	0.351	0.078	100	100	100	100
Actara	9.166	6.600	11.15	7.002	1.840	0.642	3.147	1.113
Spinosad	20.05	14.36	26.95	18.65	0.840	0.285	1.302	0.418
Malathion	205.9	75.33	345.1	83.85	0.082	0.054	0.102	0.093

Time posttreatment (hrs)

Spinosad (Tracer) than male flies. Present results indicate also that the LC<sub>50</sub> values of Spinosad are 20.1 and 16.0 ppm for males and 27.0 and 19.1 ppm for females of *B. zonata* at 24 and 48 hrs posttreatment. However, the LC<sub>50</sub> values of Spinosad recorded by El-Aw<sup>[9]</sup> against the second instar larvae of *Spodoptera littoralis* were 94.4 and 78.3 ppm at 24 and 48 hrs, respectively. It is concluded that Spinosad may be more toxic against the tephritid, *B. zonata* adults than *S. littoralis* larvae. However, Spinosad was found to be the first active ingredient in the naturally class of insect control Lepidopterous pests in cotton fields in 1997 under the name of Tracer<sup>[32]</sup>. Spinosad has been found to be highly active towards Lepidoptera<sup>[24]</sup>.

It is observed from the present results that a lower concentration of 120 ppm of Spinosad is sufficient to obtain 90 % mortality of adult flies of *B. zonata*. Jimmie and Micheal<sup>[18]</sup> demonstrated that the relative low values of Spinosad combined with a sugar-yeast hydrolysate mixture as a bait spray on female and male adults of the Caribbean fruit fly, *Anastrepha suspensa* (Loew), indicated that Spinosad was an excellent candidate for field testing.

Present results on *B. zonata* may confirm the results of El-Aw<sup>[9]</sup> who found that there was a negative relationship between the times elapsed posttreatment and the LC50 values of tested insecticides against *S. littoralis* larvae. Results of McQuate *et al.*<sup>[25]</sup> indicated that *C. capitata* was more sensitive to Spinosad than *B. cucurbitae* and *B. dorsalis*, the LC<sub>50</sub> values were 4.30, 9.16 and 9.03 ppm for all three species, respectively. Our results indicate that the LC50 values of Spinosad are 16 and 19 ppm for male and female adults of *B. zonata*, respectively. According to the present results, it is concluded that *B. zonata* is less sensitive to Spinosad than that of *C. capitata*, *B. cucurbitae* and *B. dorsalis*. However, results of McQuate *et al.*<sup>[25]</sup> in another work suggest that mass trapping with synthetic food-based bait, combined with Spinosad-based bait sprays, are control components that are compatible with biological control agent, *Fopius arisanus*, and can be combined in an integrated pest management system for *C. capitata*.

Spinosad-resistant line in a laboratory colony of *B. dorsalis* did not exhibit cross-resistance to Malathion and Methomyl<sup>[17]</sup>. Spinosad and Spinosad bait caused higher mortality to the adults of western chery fruit fly, *Rhagoletis indifferens* Curran, than Imidacloprid and Thiacloprid. Only Spinosad bait prevented oviposition<sup>[43]</sup>. However, the Spinosad-based fruit fly bait, GF120, has recently become a primary tool for area-wide suppression or eradication of tephritid fruit flies pests<sup>[42]</sup>.

Our results strongly support the finding of Nestle *et al.*<sup>[27]</sup> that the two fruit flies, *C. capitata* and *D.*

*ciliatus* were highly sensitive to low concentrations of Spinosad baited with 1 % yeast hydrolysate and 10 % sucrose as phagostimulant. The present work was carried out and achieved using *B. zonata* which is closely related to the two above-mentioned species. However, Spinosad is effective against many insects including agricultural, urban, and medical pests and was currently used in some situations for control of *Ceratitis* sp. and *Bactrocera* sp.<sup>[2]</sup>. Present results strongly indicate that Actara and Spinosad may prove safe and effective compounds as alternative to the currently used Malathion bait against PFF, *B. zonata*. Actara active ingredient, Thiamethoxam, is a second-generation neonicotinoid insecticide belonging to the thianicotinyl subclass of chemistry. This compound is registered for the control of many chewing and sucking insects such as aphids, leafminers, leafhoppers, mullein bug, plum curculio, European apple sawfly, and pear psylla. However, Actara offers new choice for fast-acting, broad spectrum insecticide that is compatible with integrated pest management programs<sup>[38]</sup>.

Spinosad is a new and highly promising insecticide with efficacy against a wide range of insects<sup>[33]</sup>. Because the contact toxicity of Spinosad is very low for both vertebrates and invertebrates and the active ingredient must be consumed in order to cause toxicity, the material is considered acceptable for use in organic agriculture<sup>[28]</sup>. Also, one of the advantages of spinosad replacement of malathion is that provides a better toxicological profile. Under experiment conditions, no spinosad residues were detected on fruit. Malathion residues were detected, although at concentrations below the maximum residue levels allowed on harvested fruits (2 ppm)<sup>[41]</sup>.

Present results on *B. zonata* support the finding of Stark *et al.*<sup>[35]</sup> who mentioned that the fruit flies; *C. capitata*, *B. cucurbitae* and *B. dorsalis* were so susceptible to Spinosad. Our results on *B. zonata* confirm again, the conclusion of many authors that Spinosad and Spinosad bait seem to be acceptable substitutes for OP and Carbamate insecticides for control tephritid fruit flies such as; Yee and Alston<sup>[43]</sup>, Xin-Geng and Russell<sup>[42]</sup>. However, because the PFF, *B. zonata* tested in this study is so susceptible to Spinosad, this product seems to be promising as a bait spray additive and replacement for Malathion for control of this species. In a further step, we will test the efficacy of Spinosad plus proteinaceous bait under commercial field conditions by comparing them against malathion bait treatment. However, in a future study the laboratory resistance and cross-resistance data may provide new information that will be useful for managing the development of resistance when spinosad is used to control *B. zonata* in the field.

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