



EVALUATION OF SYNERGISM/ANTAGONISM IN THE TOXICITY OF PYRIPROXIFEN AGAINST COTTON MEALYBUG THROUGH COADMINISTRATION WITH SILICON DIOXIDE UNDER LABORATORY CONDITIONS

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ABSTRACT

Silicon dioxide damages the protective waxy layer of insect body and insects die due to desiccation. But due to its slow action and effectiveness at high dose, its combination with different insecticides is recommended. The present study was carried out to evaluate synergistic/antagonistic effects of silicon dioxide on the toxicity of pyriproxifen against cotton mealy bug under laboratory conditions. Twenty different dose ratios between pyriproxifen and silicon dioxide ranging from 1:0.0025 to 1:3 were evaluated against adult female and egg sac stages. Two controls (one with pyriproxifen and 2nd without any treatment) were arranged for comparison purpose. The results revealed that intensity of synergistic effect on the toxicity of pyriproxifen increased with increasing concentrations of silicon dioxide. The treatments, having silicon dioxide admixed with pyriproxifen, removed many fold wax from the body of adult female than the treatments having only pyriproxifen. Approximately, 1.3 and 1.8 times greater mortality was exhibited after 1 and 2 days exposure interval than that of control, where no silicon dioxide was mixed with pyriproxifen. Similarly, 14 and 9.1 times greater wax was removed and 9.1 times greater weight loss was exhibited over control. Maximum mortality (92.5%) of nymphs at ovisac stage of mealybug was recorded, when it was exposed to ratio 1:3 and was found approximately 1.5 times greater than that of control (62.4%), where no silicon dioxide was mixed with pyriproxifen. Minimum mortality (67.5%) was observed, when it was exposed to the ratio of 1:0.0025. After a post treatment interval of 2 days, maximum mortality (94.2%) of nymphs at egg-sac stage of mealybug was observed, when it was exposed to ratio 1:3 and was at least 1.6 times greater than control (60.1%), where no silicon dioxide. The better ratio was 1:0.05 to 1:3.0. In future, silicone dioxide would be a synergist for pyriproxifen against mealybugs.

Keywords: Cotton mealybug; IGR; Laboratory bioassay; Silicon dioxide; Synergism/antagonism

INTRODUCTION

Cotton mealybug, *Phenacoccus solenopsis* (Homoptera: Pseudococcidae) is a soft-bodied insect and has emerged as an important pest of cotton in Pakistan recently within 2-3 years. Mealybugs feed on phloem tissues, suck plant sap and cause leaves to distort. The leaves become yellow and drop off and severely affected plants may die. This insect also produces honey dew resulting in sooty mold growth, which hinders photosynthesis process (Saeed *et al.*, 2007). In Pakistan, cotton mealy bug has been tried to be controlled by application of insecticides in form of cover spray (Saeed *et al.*, 2007) which causes significant mortality in initial instars that

lack waxy covering; but non significant (nominal) mortality in later instars and adult females as they have very thick waxy covering. Similarly, eggs/nymphs inside the egg sac remain intact from the exposure of insecticides and survive successfully (Curkovic *et al.*, 2007). It is therefore, imperative to evaluate any wax dissolving agent which synergizes the toxicity of insecticides by enhancing the penetration of insecticide. Diatomaceous earth has long been known as a potentially useful insecticide. It is composed almost entirely of amorphous silicon dioxide, formed from fossilized diatoms known as single-celled algae (Quarles, 1992; Ziaee *et al.*, 2006). It has been very effective because it damages the waxy layer of insect body and insect die due to

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dehydration (Korunic, 1998; Subramanyam and Roesli, 2000). Silicon dioxide, in combination with different insecticides, has been evaluated against many stored insect pests (Riasat *et al.*, 2011; Wakil *et al.*, 2010, 2012, 2013). This study was carried out to evaluate the synergistic/antagonistic effects of silicon dioxide on the toxicity of an IGR, pyriproxifen against cotton mealybug (field crop insects) under laboratory conditions. The main hypotheses evaluated through this study were: either silicon dioxide synergizes/antagonizes the toxicity of pyriproxifen against cotton mealybug, silicon dioxide removes the waxy covering from the body of cotton mealybug and dissolves the waxy fluff of mealybug egg sac or silicon dioxide leaves blotches on cotton leaves or not.

MATERIALS AND METHODS

Pyriproxifen was coadministered with silicon dioxide at their recommended doses for the evaluation of their toxicity against cotton mealy bug under laboratory conditions. Silicon dioxide (SiO₂: M=60.08, granular form) was purchased and its one molar solution was prepared in diluted hydrofluoric acid under laboratory conditions (32 ± 1°C and 65% RH). This one molar solution, then, was used to prepare test solution. Pyriproxifen at its recommended dose was coadministered with one molar solution of silicon dioxide in twenty different ratios i.e., 1:0.0025, 1:0.005, 1:0.0075, 1:0.001, 1:0.025, 1:0.05, 1:0.075, 1:0.01, 1:0.25, 1:0.5, 1:0.75, 1:1, 1:1.25, 1:1.5, 1:1.75, 1:2, 1:2.25, 1:2.5, 1:2.75 and 1:3 (pyriproxifen: Silicon dioxide) with the help of pipette in separate beakers. The test solutions were applied on mature females and eggsac with the help of atomizer. The data regarding mortality of each stage was recorded 1 day and 2 days post treatment application. After each observation, the dead adult females as well as stage inside the eggsac (1st instar nymph) were counted and removed from the treated petridish. They were released in separate petridish with fresh food to check the original number of died insects. The data collected were transformed into percent corrected mortality by abbot formula (Abbot, 1925). The nymphs emerging from treated and untreated (control) were counted with the help of microscope and %age mortality at egg sac stage was calculated. The adult female was also weighed before and an hour after treatment application to measure the quantity of wax removed. The mortality as well as wax data were subjected to ANOVA technique to evaluate the variation in the toxicity of test solution as well as in the quantity of wax removed; whereas the means of test solutions (treatments) with significant differences were subjected to Tukey HSD (Honestly Significant Difference) test for comparison (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Synergistic effects against adult female

Maximum mortality (93.3%) of adult mealybug was recorded, when it was exposed to ratios ranging from 1:0.5 to 1:3 (recommended dose of pyriproxifen: silicon dioxide concentration) for one day, that was at least 1.8 times greater than that of control (53.3%), where no silicon dioxide was

mixed with pyriproxifen. Minimum mortality (66.7%) was observed, when adult female was exposed to the ratios 1:0.0025 and 1:0.005 (Table-1). At post treatment interval of 2 days, maximum mortality (93.3%) of adult mealybug was observed, when it was exposed to ratios ranged from 1:0.05 to 1:3 and was at least 1.3 times greater than that of control (73.3%), where no silicon dioxide was mixed with pyriproxifen. Minimum mortality (86.7%) was observed, when adult female was exposed to the ratios ranging from 1:0.0025 to 1:0.025 (Table-2). The percentage mortality of adult female mealybug for all other ratios increased with increasing the concentrations of silicon dioxide admixed with pyriproxifen. A little literature on the efficacy of pyriproxifen in combination with silicon dioxide is available. For example, El-Wakeil and Saleh (2009) showed that diatomaceous earth increased the efficacy of neemazal, when used in combination against *Myzus persicae* (Sulzer) as compared to use alone.

Maximum quantity of wax (4.2 mg) was removed from the body of adult mealybug, when it was exposed to a ratio 1:3 (recommended dose of pyriproxifen: silicon dioxide concentration) for 1 hr, that was approximately 14 times greater than that of control (0.3 mg), where no silicon dioxide was mixed with pyriproxifen. Minimum quantity of wax (0.6 mg) was removed from the body of adult mealybug when it was treated with the ratio 1:0.0025 (Table-1). The quantity of wax removed from the body of adult female mealybug for all other ratios increased with increasing the concentrations of silicon dioxide admixed with pyriproxifen. Maximum weight loss (64.6%) of adult mealybug was recorded, when it was exposed to ratio 1:3 (recommended dose of pyriproxifen: silicon dioxide concentration) for 1hr, that was approximately 9.1 times greater than that of control (7.1%), where no silicon dioxide was mixed with pyriproxifen. Minimum weight loss (12.1%) was observed, when adult female was exposed to the ratio 1:0.0025 (Table-1). The percentage weight loss of adult mealybug for all other ratios increased with increasing the concentrations of silicon dioxide admixed with pyriproxifen. No information regarding synergistic effects of silicon dioxide on pyriproxifen or other insecticides against mealybug was available; so it cannot be compared or contradicted with the present results. The enhanced mortality in adult mealybug by pyriproxifen, mixed with silicon dioxide, was attributed to its wax dissolving property because adult females treated with silicon dioxide mixed pyriproxifen exhibited less wax on their body and more reduction in their body loss.

Synergistic effects against nymphs at ovisac stage

Maximum mortality (92.5%) of nymphs at ovisac stage of mealybug was recorded, when it was exposed to ratio 1:3 (recommended dose of pyriproxifen: silicon dioxide concentration) for one day and was found approximately 1.5 times greater than that of control (62.4%), where no silicon dioxide was mixed with pyriproxifen. Minimum mortality (67.5%) was observed, when it was exposed to the ratio of 1:0.0025 (Table-1). After a post treatment interval of 2 days, maximum mortality (94.2%) of nymphs at egg-sac stage of mealybug was observed, when it was exposed to ratio 1:3 and was at least 1.6 times greater than control (60.1%), where no silicon dioxide was mixed with pyriproxifen. Minimum

mortality (69.4%) was observed, when it was exposed to a ratio 1:0.0025 (Table- 2). The percentage mortality at egg-sac stage of mealybug for all other ratios increased with increasing the concentrations of silicon dioxide admixed with pyriproxifen. The results are in agreement with those of Lord

(2001), Akbar *et al.* (2004) and Wakil *et al.* (2011) who reported that diatomaceous earth increased the efficacy of *Beauveria bassiana*, an entomopathogenic fungus, when used in combination against insects pest as compared to use alone.

Table 1

Percentage mortality of adult female and nymphs at ovisac stage, quantity of wax removed and % weight loss (Means \pm S.E.) 1 hr post treatment exposure to different ratios between pyriproxifen and silicon dioxide.

Ratio (pyriproxifen silicon dioxide)	Mortality of adult Female (%)	Wax removed (mg)	Weight loss (%)	Mortality at ovisac Stage (%)
1:0.0025	66.7 bc \pm 13.2	0.6 klm \pm 0.06	12.1 ghi \pm 1.2	67.5 m \pm 0.23
1:0.005	66.7 bc \pm 13.3	0.7 jklm \pm 0.07	12.4 gh \pm 2.9	70.3 l \pm 0.24
1:0.0075	73.3 abc \pm 6.4	0.8 ijkl \pm 0.01	17.1 gh \pm 1.6	71.2 kl \pm 0.24
1:0.01	73.3 abc \pm 5.2	0.8 ijkl \pm 0.09	17.4 g \pm 1.3	71.9 k \pm 0.25
1:0.025	73.3 abc \pm 6.3	1.2 hijk \pm 0.1	18.1 g \pm 1.2	74.2 j \pm 0.26
1:0.05	73.3 abc \pm 5.3	1.3 ghij \pm 0.9	20.0 fg \pm 2.3	74.5 j \pm 0.28
1:0.075	80.0 ab \pm 20.0	1.4 fghi \pm 0.3	28.5 ef \pm 2.6	76.4 i \pm 0.27
1:0.1	80.0 ab \pm 19.2	1.5 fgh \pm 0.08	31.1 e \pm 1.7	80.1 h \pm 0.29
1:0.25	86.7 ab \pm 12.2	1.9 efg \pm 0.8	34.0 de \pm 2.4	80.4 gh \pm 0.22
1:0.5	93.3 a \pm 4.9	2 def \pm 0.2	36.0 cde \pm 1.9	80.8 gh \pm 0.21
1:0.75	93.3 a \pm 3.9	2.0 def \pm 0.4	41.8 bcd \pm 2.0	81.6 fg \pm 0.30
1:1	93.3 a \pm 6.4	2.2 cde \pm 0.5	42.5 bcd \pm 1.8	83.0 ef \pm 0.31
1:1.25	93.3 a \pm 5.9	2.4 bcde \pm 0.7	43.6 bcd \pm 1.1	83.4 e \pm 0.29
1:1.5	93.3 a \pm 6.5	2.4 bcde \pm 1.3	43.9 bcd \pm 2.2	83.6 de \pm 0.36
1:1.75	93.3 a \pm 5.5	2.6 bcd \pm 1.4	44.2 bc \pm 2.6	84.1 de \pm 0.28
1:2	93.3 a \pm 4.8	2.7 bc \pm 0.9	44.8 bc \pm 2.7	85.0 d \pm 0.29
1:2.25	93.3 a \pm 6.3	2.7 bc \pm 1.6	49.1 b \pm 1.4	86.4 c \pm 0.37
1:2.5	93.3 a \pm 6.7	2.9 b \pm 1.4	50.0 b \pm 2.5	87.3 bc \pm 0.32
1:2.75	93.3 a \pm 6.1	4.0 a \pm 1.3	51.1 b \pm 2.4	88.2 b \pm 0.30
1:3	93.3 a \pm 5.6	4.2 a \pm 2.1	64.6 a \pm 3.1	92.5 a \pm 0.31
0:0	0.0 d \pm 0.0	0.0 m \pm 0.0	0.0 i \pm 0.0	0.0 o \pm 0.0
1:0	53.3 c \pm 6.2	0.3 lm \pm 0.01	7.1 hi \pm 0.6	62.4 n \pm 0.22

Table-2

Percentage mortality in adult female and at ovisac stage (Means \pm S.E.) 1 day post treatment exposure to different ratios between pyriproxifen and silicon dioxide.

Ratio (pyriproxifen: silicon dioxide)	Mortality of adult female (%)	Mortality at ovisac Stage (%)
1:0.0025	86.7 ab \pm 13.3	69.4 k \pm 0.34
1:0.005	86.7 ab \pm 13.0	72.2 j \pm 0.35
1:0.0075	86.7 ab \pm 13.1	73.1 j \pm 0.33
1:0.01	86.7 ab \pm 12.9	73.8 j \pm 0.34
1:0.025	86.7 ab \pm 13.2	76.1 i \pm 0.35
1:0.05	93.3 a \pm 6.5	76.4 i \pm 0.36
1:0.075	93.3 a \pm 6.6	78.4 h \pm 0.37
1:0.1	93.3 a \pm 6.7	82.0 g \pm 0.34
1:0.25	93.3 a \pm 5.6	82.3 g \pm 0.35
1:0.5	93.3 a \pm 5.7	82.6 g \pm 0.34
1:0.75	93.3 a \pm 5.5	83.5 fg \pm 0.39
1:1	93.3 a \pm 6.4	84.9 ef \pm 0.31
1:1.25	93.3 a \pm 6.3	85.3 def \pm 0.32
1:1.5	93.3 a \pm 5.4	85.5 de \pm 0.41
1:1.75	93.3 a \pm 5.3	85.9 de \pm 0.43
1:2	93.3 a \pm 4.9	86.8 cd \pm 0.42
1:2.25	93.3 a \pm 4.8	88.2 bc \pm 0.48
1:2.5	93.3 a \pm 3.9	89.3 b \pm 0.40
1:2.75	93.3 a \pm 6.0	90.0 b \pm 0.44
1:3	93.3 a \pm 6.1	94.2 a \pm 0.34
0:0	0.0 c \pm 0.0	0.0 m \pm 0.0
1:0	73.3 b \pm 0.2	60.1 l \pm 0.36

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