



## **Investigating the Effects of a Mixture of Diatomaceous Earth and Spinosad Insecticide to Control Adult Flour Weevils, *Tribolium castaneum* Herbst. (Col: Tenebrionidae)**

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### **ABSTRACT**

The red flour weevil, *Tribolium castaneum* Herbst, is one the major pests of stored products. Adult pests and larvae of this pest causes economic damage through releasing toxic substances into flour. Larvae shells and adult pests affect the quality of the flour. The effectiveness of spinosad and diatomaceous earth insecticides on adult og this pests was studied. The amount of LC50 diatomaceous earth with silico-sec® formulation for adult insects of the red flour weevil and Spinosad insecticide was estimated to be 2.34 g/m<sup>2</sup> and 76.11 mg/l, respectively. After 14 days, the mortality rate was 100%. The results showed that a mixing of diatomaceous earth and Spinosad an increase in the efficacy of the mixture in causing loss. In addition, with increasing time of exposure and dose mortality rate increases.

**Keywords:** Flour Weevil, Diatomaceous Earth, Spinosad, silico-sec®.

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Cumulative mode occurs when the toxicity of the mixture of the two compounds is approximately equivalent to the sum of their components (Talebi Jahromi, 2007).

### **1. INTRODUCTION**

The red flour weevil, *Tribolium castaneum* Herbst, is one pests of stored products, especially flour, that causes economic damage of it (Songa & Rono, 1998).

The use of non-chemical methods, such as natural, biological, and microbial control methods against storage pests, has attracted the attention of researchers as a result of the harm done by chemical insecticides (Doostdar kolkenari and colleagues, 2015).

Methyl bromide and phosphine are used to control pest infestations, but they have a severe toxicity for humans. (Ayvaz et al., 2010).

Diatomaceous Earth(DE) is a type of biocompatible compounds used to protect grains and legumes in storehouses. Based on the reports of International Agency for Research on Cancer Diatomaceous Earth , belongs to the third generation of pesticides and it isn't carcinogenic. (Armitage et al, 1998).

Recent studies have suggested that biological spinosad insecticides are effective against a wide range of pest infestations and have fatal effects. (Thomson et al.,2000).

Spinosad insecticides are low in nature and are relatively non-toxic to mammals, birds and fishes (Talebi Jahromi, 2007).

Synergism occurs when both compounds are used together or sequentially, and their toxicity intensifies when used alone. The antagonistic phenomenon is the opposite of the phenomenon of synergism, and it happens when the mixture of the two compounds reduces their insecticidal effects.

### **2. MATERIALS AND METHODS**

#### **2.1. Breeding Insects**

To conduct tests of this research, samples of the pest of *T. Castaneum* were supplied from Entomology Laboratory of the University of Maragheh. Insect breeding cube was used in the form of plastic container .Red weevil was bred in laboratory in 28±1 centigrade temperatures, 55±5% relative humidity darkness. Culture environment containing 95% whole wheat flour and 5% barley malt (Golestan hashemi, 2011).

The insects were reared for six generations in the laboratory and were used for biometric tests.

#### **2.2. Biometric tests**

##### **Determining LC<sub>50</sub>**

Initial bioassay experiments were performed to calculate the 50% lethal concentration (LC<sub>50</sub>) of the biocompatible spinosad insecticide.Then to determine to determine the lowest concentration and the highest concentrations required for the test including 150,112, 83, 60 and 50 (mg/lit) for the flour weevil along with the control treatment, the experiments were performed using petri dish method. The experiment had 3 replications and there were 20 insects for each replicate. Also, in order to determine LC<sub>50</sub> of Diatomaceous Earth(DE), preliminary experiments were performed. Glass petri dishes with 9 cm inner diameter and 63/5 cm<sup>2</sup>inner area were used. In order to perform this experiment, (1,3,4,6,8) g/m<sup>2</sup>doses of Diatomaceous Earth along with control treatment group were selected. The experiment was performed in a complete

randomized design with 3 replications. 3 ml distilled water was added to each of the doses to make particles of diatomaceous earth stick to the inner surface of petri dishes. After the water on the inner surface of the petri dishes evaporated, ten 3 to 4-day old adult flour weevils were released. Next, after 1 to 7-day periods, the number of deaths was counted and recorded.

The doses used in this experiment, after the preliminary experiment, including (6,5,4,3,5, and 3) g/m<sup>2</sup> Diatomaceous Earth along with control treatment group were selected. This experiment was performed in a complete randomized design with 3 replications, too. The containers were kept in 27 ± 1 temperature and 60 ± 5 relative humidity. After 1 to 7-day periods, the mortality rate of the insects was registered. The effect of mixing diatomaceous compounds and spinosad insecticide compounds was performed based on the results of previous experiments. To perform these experiments, after determining LC<sub>50</sub> +LC<sub>30</sub>, both Diatomaceous Earth and Spinosad insecticide, the 76.11 and 51.58 mg/lit concentrations from the mixture of Spinosad and 4.24 and 3.42 g/m<sup>2</sup> doses of Diatomaceous Earth were selected.

### 2.3. Analysis of data

The data obtained from various experiments of this study were analyzed using SAS statistics software. Probit method in SAS software was used to determine the values of LC<sub>30</sub>, LC<sub>50</sub>, and LC<sub>90</sub>. LSD test was used to compare the means. Microsoft excel worksheet was used to draw diagrams.

## 3. RESULTS AND DISCUSSION

The results and analysis of the relevant data from biometric experiments of Spinosad on flour weevil are presented in Table 1. LC<sub>50</sub> and LC<sub>90</sub> values were 76.11 and 196.91 mg/l, respectively. The results indicate that if concentrations increases, the mortality rate will also increase.

**Table 1:** Spinosad's Toxicity on Adult Flour Weevils after 24 Hours

| Insecticides | No. | X <sup>2</sup> | Line slope | Lethal Concentrations( mg/l) |                  |                  |
|--------------|-----|----------------|------------|------------------------------|------------------|------------------|
|              |     |                |            | LC <sub>30</sub>             | LC <sub>50</sub> | LC <sub>90</sub> |
|              |     |                |            | Dose-effect<br>(95%FL)       | (95%FL)          | (95%FL)          |
| Spinosad     | 360 | 41/57          | 3/10 ± ·4  | 51/58                        | 76/1             | 196/91           |

**Table 2:** Toxicity Rate of silico-sec® Formulation of DE on Adult Flour Weevil

| Insecticides | No. | X <sup>2</sup> | Line slope | Lethal doses( g/m <sup>2</sup> ) |                  |                  |
|--------------|-----|----------------|------------|----------------------------------|------------------|------------------|
|              |     |                |            | LC <sub>30</sub>                 | LC <sub>50</sub> | LC <sub>90</sub> |
|              |     |                |            | Dose-effect<br>(95%FL)           | (95%FL)          | (95%FL)          |
| DE           | 360 | 56/15          | 5/64 ± ·7  | 3/42                             | 4/24             | 7/15             |

Based on the data obtained from this study, the increase in the formulation of DE leads to an increase in the mortality of flour weevils (Table 2).

This finding is consistent with the observations of Ulrichs and Mewis (2001) experiments. In this investigation on the mortality of DE formulation of Fossil Shield® on *Tribolium confusum* and *Tenebrio molitor* They found that after 7 days,

decreases the number of adult insects and after 14 days it causes 100% mortality.

The data obtained from comparing the percentages of mortality through using Duncan's Multiple Range Test with 95 % reliability shows that mixture of Spinosad and DE in LC<sub>50(DE)</sub> +LC<sub>30(SP)</sub> concentrations caused the highest rate of mortality in one-day old adult *T. castaneum*. The effects of mixing Spinosad insecticide and DE in this experiment is clearly observable.

**Table 3:** Toxicity Rate of silico-sec® Formulation of DE on Adult Flour Weevil

| No. | Treatment                                  | Mortality Mean | Grouping |
|-----|--|----------------|----------|
| 1   | LC <sub>50(DE)</sub> +LC <sub>50(SP)</sub> | 100 0± /00     | a        |
| 2   | LC <sub>50(SP)</sub> +LC <sub>30(DE)</sub> | 86/66 ± 1/66   | b        |
| 3   | LC <sub>50(DE)</sub> +LC <sub>30(SP)</sub> | 78/33 ± 1/66   | c        |
| 4   | LC <sub>30(DE)</sub> +LC <sub>30(SP)</sub> | 68/33 ± 1/66   | d        |

Different letters indicate the significant discrepancy at the 5% level of statistical probability in Duncan's test.

The results obtained from this study on mixing Diatomaceous Earth and Spinosad shows that adding Diatomaceous Earth to Spinosad insecticide causes an increase in the fatality rate. In laboratory studies mortality rate of *T. confusum* in the wheat treated with concentration 1000 ppm of Spinosad after using DE increase. ( Chintzoglou et al., 2008).

It's been proven that spinosad insecticide can be used as an appropriate alternative for chemical control. It is also recommended to be used in conjunction with other pest control methods to protect crop products. In addition, the application of diatomaceous earth mixtures with spinosad reduces the concentration of spinosad and increases its effect. (Golestan hashemi, 2011).

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