

(RESEARCH ARTICLE)



## Toxic and repellent potentials of spinosad against *Cryptolestes pusillus* (Schon.) (Coleoptera: Cucujidae)

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### Abstract

The contact and gustatory effects of spinosad on the mortality and repellency of *Cryptolestes pusillus* (Schon.) were conducted under laboratory conditions. Spinosad at 0.32 l/g caused lowest mortality ( $16.67 \pm 1.67\%$  of 2<sup>nd</sup> instar larva of *C. pusillus* in wheat after 24h and highest mortality ( $59.14 \pm 1.65\%$ ) was observed at 1.25 l/g in wheat after 72 h exposure. The  $LC_{50}$  value was 0.11 l/g in wheat after 72 h, which indicated that spinosad is highly toxic against the 2<sup>nd</sup> instar larvae of *C. pusillus*. In case of 4<sup>th</sup> instar larvae highest mortality ( $58.12 \pm 3.45\%$ ) were observed at 1.25 l/g concentration but lowest  $10.00 \pm 2.10$  after 24 h exposure. The highest mortality was observed ( $40.00 \pm 2.88\%$ ) in adults. The different doses (1.25, 0.63, 0.32, 0.16 and 0.08 l/ml) of spinosad showed repellent activities against adults of *C. pusillus*. All the doses of spinosad offered 0.1% level of significance ( $P < 0.01$ ). Spinosad used in this experiment have great potential in the control of *C. pusillus* which is important from the Integrated Pest Management and the Global Environmental Protection point of views.

**Keywords:** Toxicity; Repellent; Spinosad; *Cryptolestes pusillus*; Wheat

### 1. Introduction

Insect pest management in stored food commodities using chemicals is facing many challenges due to the concerns of human health safety, development of insect resistance against the chemical pesticides and creating environmental hazards (Nayak *et al.* 2005, Daghli and Nayak 2006). The future food security of a nation is a major threat of Insect attack of stored grain loss in quality and quantity. We are already a large population country now. Whole world is going on a common problem of vast population and food frugality. At the beginning of the 21<sup>st</sup> century, we are faced a great problem of food crisis. It's a dehumanizing loopholes of any nation. As a developing country Bangladesh is alarming position in total population and food management. Approximately 25 percent of the population in Bangladesh remains food-insecure and 36 percent of children younger than five years of age suffer from stunting, a common measure of chronic malnutrition (WFP 2020).

Wheat (*Triticum aestivum* L.) is the second most important staple food after rice that accounts for about 12 percent of total cereal consumption. Several insect pests are damaged large number of stored wheat. Increasing global temperature per/degree researchers opinion, coleopteran insect one of them are damaged beyond 10-25% (BBC NEWS 2018). *Cryptolestes pusillus* is a coleopteran pest which destroy stored grain. It is commonly known as flat grain beetle and belongs to family Cucujidae under the order Coleoptera. The beetle is an external feeder and a serious cosmopolitan pest of stored product commodities especially cracked grains (Barker 1976). It multiplies rapidly and subsequently build up into a huge population within very short period of time (Rahman *et al.* 2009). It does not attack the whole grain but feeds upon broken kernels and the dust resulting from attacks of other grain-feeding insects (Pest Management

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Spinosad, a reduced-risk commercial insecticide derived from a bacterial fermentation product, possesses both contact and oral toxicities against insects. Previous studies document that the insecticidal efficacy of spinosad is affected by several biotic or abiotic factors, such as the target species, the type of commodity, the exposure interval and the type of surface that spinosad is applied to (Toews and Subramanyam 2003, Daghli and Nayak 2006, Subramanyam 2006). So far, spinosad has proved to be very effective against a wide range of stored-product pests, and can retain its efficacy for a long time after application. Daghli and Nayak (2006) and Subramanyam *et al.* (2007) found that in stored maize, spinosad remained stable for a two-year period. Spinosad is registered as a grain protectant in the USA at the labeled rate of 1 ppm (Subramanyam 2006) and is expected that registration for purpose will be expanded in other parts of the world. Agro Sciences (Indianapolis, Indiana, USA) based on chemical compounds of a soil bacterium *Saccharopolyspora spinosa* was discovered in 1985 (Mertz and Yao 1990). It is a naturally derived bio-rational insecticide with an environmentally favorable toxicity profile (Bond *et al.* 2004).

The present investigation was designed to evaluate the toxicity and repellent potential of spinosad against *C. pusillus* larvae and adults under laboratory conditions.

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## 2. Material and methods

### 2.1. Methods of the bioassay

In the present study exposure of different life stage of *C. pusillus* and were exposed to treated Food Method (TEM) (Talukder and Howse 1994) for evaluating the effects of spinosad against the larvae and adults of the beetle.

#### 2.1.1. Collection of *C. pusillus*

*C. pusillus* beetles were obtained from the stock culture of laboratory without any exposure to insecticides, maintained in the control temperature (CT) room at Entomology and Insect Biotechnology laboratory, Institute of Biological Sciences, University of Rajshahi, Bangladesh.

#### 2.1.2. Preparation of standard food medium for mass culture of *C. pusillus*

Wheat collected from Wheat Research Institute, Shampur, Rajshahi, Bangladesh. These grains were washed with water and dried at room temperature before adjusting their moisture content to 13.5 by adding tap water. Then cleaned by sieving through 500 micrometer aperture sieve and sterilized in an oven at 60°C for 6h. After sterilization wheat grains were kept in different plastic containers that were cleaned before. Some grains were partially broken down by the hand blender to use throughout the experimental period for *C. pusillus*.

#### 2.1.3. Source of Spinosad

About 500ml of liquid spinosad (PRN- MAPP-12054, cfno 20012- 019, lot No- 3068404) was obtained from Dow Agro Sciences, UK. The liquid is light grey to white in colour with slight odour stale water. Concentration of spinosad was 120g spinosad/ Litre.

#### 2.1.4. Preparation of spinosad concentrations

The spinosad was diluted in distilled water. In 50ml beaker 8.75µl spinosad was taken by using a micropipette, and 3ml distilled water were added properly in it by using 2ml & 1ml syringe (1 time). The vial was shaken vigorously for equal mixing of spinosad and water. From this solution 1ml was taken off which contained 1.25µ/ml spinosad, which was the stock concentration. The other concentrations of spinosad were prepared by serial dilution of stock solution and adding 2ml distilled water in each step. So, the desired concentrations of spinosad were obtained as 0.30, 0.63 and 1.25, which were used for toxicity study against *C. pusillus*.

#### 2.1.5. Commodities used

Untreated and infestation free hard red wheat variety was used in the experiments. The grains were washed with water and dried at room temperature before adjusting their moisture content to 13.5% by adding tap water. The grain was sieved through 500 micrometer aperture sieve and sterilized in an oven at 100°C for 8h. After sterilization wheat grain was broken down by the hand blender and kept in clean plastic containers for using throughout the experimental period.

### 2.1.6. Bioassays

This cracked wheat was used as food medium for *C. pusillus* larvae and adults. One gram (1gm) of wheat grain of a variety was placed in a petri-dish (6cm) and treated with freshly prepared aqueous spinosad solution of a definite concentration using a 1ml syringe. Ten pairs of either 2<sup>nd</sup> or 4<sup>th</sup> instar larvae of *C. pusillus* was released in the treated wheat separately. Similarly, 10 pairs of untreated adults (3-5d old) were also released separately in the treated wheat. The petri-dish were covered with lid and kept in the CT room at 30±1C and 75±0.5C% RH. Mortality of the larval instars and adults of *C. pusillus* were recorded after 24, 48 and 72h exposure periods. For control batch wheat grain were treated with 1micro liter distilled water only. Three replications were taken for each of the spinosad concentrations, wheat variety each larval instar and adults of *C. pusillus*.

### 2.2. Data collection and statistical analysis

The data for percent kill of *C. pusillus* in all used concentration was recorded after 24, 48 and 72h exposure; the data were subjected to Analysis of Variance using ANOVA. Means were compared by Tukey's tests ( $P < 0.05$ ), and subjected to probit Analysis using the probit software for calculating average larval and adult mortality data and estimation of LC<sub>50</sub> (lethal concentration) values. The regression lines were drawn using Microsoft Excel-2010 and Bio Stat-2009. The percent reduction of adult emergence in treatments compared to control (PRC) was calculated by using the formula provided by Main and Mulla (1982) as follows:

$$\text{PRC} = 1 - \{ \text{Average no. of adult emergence (treatment)} / \text{Average no. of adult emergence control} \} \times 100$$

The mortality record was corrected by the Abbott's (1925) formula in the following manner:

$$P_r = \{ (P_o - P_c) / (100 - P_c) \} \times 100$$

Where,  $P_r$  = Corrected mortality (%)

$P_o$  = Observed mortality (%)

$P_c$  = Control mortality (%), sometimes called natural mortality (%).

### 2.3. Application of doses for repellency of *C. pusillus*

The repellency test was adopted from the method of Talukder and Howse (1994). Half filter paper disc (Whatman n. 40, 9 cm diam) were prepared and selected doses were applied onto each of the half-disc was then attached lengthwise, edge-to-edge, to a control half-disc with adhesive tape and placed in a Petri dish (9 cm diam), the inner surface of which was smeared with flu on to prevent insect escaping. The orientation of the stimulus affecting the distribution of the test insects. Ten adult insects were released in the middle of each filter-paper circle. Each concentration was tested five times. Insects that settled on each half of the filter paper discs were counted after 1 h and then at hourly intervals for 5 h. The average of the counts was converted to percentage repellency ( $PR$ ) using the formula of Talukder and Howse (1995):

$$PR = 2 (C - 50),$$

Where,  $C$  is the percentage of insects on the untreated half of the disc. Positive values expressed repellency and negative values for attractant.

Repellency was observed for one-hour interval and up to five successive h of exposure, just by counting the number of insects in the treated and non-treated part of the filter paper spread on the floor of the 90 mm Petri dish. The values in the recorded data were then calculated for percent repellency and then ANOVA.

## 3. Results and discussion

### 3.1. Toxicity of spinosad against 2<sup>nd</sup> instar larva

The lethal result of spinosad on 2<sup>nd</sup> instar larvae of *C. pusillus* in wheat at different concentrations and different exposure periods are shown in Table 1. The result showed that when the 2<sup>nd</sup> instar larvae were exposed to spinosad treated wheat at various concentrations (0.32, 0.63 and 1.25µl/g) for different time interval (24, 48 and 72h) the toxic effects of the spinosad was differed with different exposure periods. Spinosad at 0.32µl/g caused lowest mortality (16.67±1.67%) of 2<sup>nd</sup> instar larva of *C. pusillus* in wheat after 24h, and highest mortality (59.14±1.65%) was observed at 1.25µl/g in wheat after 72h exposure. The out put was positively related to concentration of spinosad and exposure time of the larvae. The LC<sub>50</sub> values, 95% Confidence limits, regression equations,  $\chi^2$  for heterogeneity and regression lines of empirical probit

mortality of 2<sup>nd</sup> instar larvae of *C. pusillus* are presented in Table 2. The LC<sub>50</sub> values clearly show that spinosad toxicity was increased with the increase of exposure periods. The lowest LC<sub>50</sub> value was 0.11 µl/g in wheat after 72h, which indicated that spinosad is highly toxic against the 2<sup>nd</sup> instar larvae of *C. pusillus*.

ANOVA results clearly indicated highly significant interaction effects were present between spinosad concentrations (F= 786.52, df=2 P<0.001) and exposure time (F= 335.60, df=2, P<0.001) (Table 1).

### 3.2. Toxicity of spinosad against 4<sup>th</sup> instar larva

The lethal result of spinosad on 4<sup>th</sup> instar larvae of *C. pusillus* in wheat at different concentrations and different exposure periods are shown in Table 3. Highest mortality of 4<sup>th</sup> instar larvae was noted as 58.12±3.45% at 1.25 µl/g concentration and 72h exposure, and lowest mortality was 10.00±2.10% after 24h exposure at the same concentration. Spinosad mortality against the 4<sup>th</sup> instar larvae of *C. pusillus* was positively related with both its concentrations and exposure period, which were significantly different between the control and treatments.

The LC<sub>50</sub> values, 95% confidence limits, regression equations,  $\chi^2$  for heterogeneity and regression lines of empirical probit mortality of 4<sup>th</sup> instar larvae are shown in Table 4. Toxic effect was maximum (LC<sub>50</sub> 0.749) at 72h exposure. ANOVA results clearly expressed high significant interaction effects were found between spinosad concentrations (F=886.82, df=2, P<0.001) and exposure periods (F=308.94, df=2, P<0.001) (Table 3).

### 3.3. Toxicity of spinosad in unsexed adults

**Table 1** Toxicity of different concentrations of spinosad against 2<sup>nd</sup> instar larvae *C. pusillus* after 24, 48 and 72h of exposure

Concentrations (µl/g)	Average Mortality %		
	Exposure period (h)		
	24	48	72
Control	0.00±0.00d	0.00±0.00d	0.00±0.00d
0.32	8.56±1.11c	20.22±1.1c	34.33±2.24c
0.63	14.54±1.1b	30.00±1.68b	40.00±2.26b
1.25	22.12±2.00a	34.00±2.12a	54.12±1.65a

In a column means with same letter do not significantly differed from each other with in Concentrations at 0.05% level (Tukey,s test)

### ANOVA

Source	DF	F value
Concentrations	2	786.52***
Exposure time	2	335.60***
Total	4	

In a column means with same letter do not significantly differed from each other within varieties at 0.05% level (Tukey's test). Note: \*\*\*= Significant at P<0.001, NS= Non Significant

**Table 2**  $\chi^2$  for heterogeneity, regression equations, LC<sub>50</sub> and 95% confidence limits of different concentrations of spinosad against 2<sup>nd</sup> instar larvae *C. pusillus* after 24, 48 and 72 h exposure time

Exposure time (h)	$\chi^2$ for heterogeneity	Regression equation	LC <sub>50</sub> (µl g <sup>-1</sup> )	95% confidence limits	
				Lower	Upper
24	0.065	Y=3.072258+0.4590699X	1.011	0.023	36.68
48	0.0004	Y=3.392423+0.5044368X	0.120	0.031	0.60
72	0.36	Y=3.767731+0.8220564X	0.011	0.006	0.038

Results of adult mortality with different concentrations of spinosad after different concentrations of spinosad after different exposure periods are presented in Table 5. The highest concentration of spinosad 1.25 $\mu\text{l/g}$  at 72h resulted in to  $\geq 50\%$  mortality in wheat in all concentrations and exposure periods. The highest mortality was observed (40.00 $\pm$ 2.88%).

**Table 3** Toxicity of different concentrations of spinosad against 4<sup>th</sup> instar larvae of *C. pusillus* after 24, 48 and 72h of exposure

Concentrations ( $\mu\text{l/g}$ )	Average Mortality %		
	Exposure period (h)		
	24	48	72
Control	0.00 $\pm$ 0.00d	0.00 $\pm$ 0.00d	0.00 $\pm$ 0.00d
0.32	10.00 $\pm$ 2.0c	18.48 $\pm$ 1.8c	26.68 $\pm$ 2.10c
0.63	18.33 $\pm$ 2.4b	25.00 $\pm$ 1.68b	33.32 $\pm$ 2.22b
1.25	24.00 $\pm$ 2.25a	31.15.00 $\pm$ 2.21a	58.12 $\pm$ 3.68a

In a column means with same letter do not significantly differed from each other within varieties at 0.05% level (Tukey's test)

#### ANOVA

Source	DF	F value
Concentrations	2	886.83***
Exposure time	2	308.94***
Total	4	

In a column means with same letter do not significantly differed from each other within varieties at 0.05% level (Tukey's test). Note: \*\*\*= Significant at  $P < 0.001$ , NS= Non Significant

**Table 4**  $\chi^2$  for heterogeneity, regression equations, LC<sub>50</sub> and 5% confidence limits of different concentrations of spinosad against 4<sup>th</sup> instar larvae of *C. pusillus* after 24, 48 and 72h exposure period

Exposure time (h)	$\chi^2$ for heterogeneity	Regression equation	LC <sub>50</sub> ( $\mu\text{l g}^{-1}$ )	95% confidence limits	
				Lower	Upper
24	0.003	Y=2.107791+1.115524X	3.922	1.480	8.417
48	0.768	Y=2.683544+0.9026346X	2.563	1.124	4.053
72	0.005	Y=2.736731+1.209109X	1.112	0.374	1.115

**Table 5** Toxicity of different concentrations of spinosad against adult (unsexed) *C. pusillus* after 24, 48 and 72h of exposure

Concentrations ( $\mu\text{l/g}$ )	Average Mortality %		
	Exposure period (h)		
	24	48	72
Control	0.00 $\pm$ 0.00d	0.00 $\pm$ 0.00d	0.00 $\pm$ 0.00d
0.32	8.67 $\pm$ 1.67c	17.83 $\pm$ 1.67c	26.33 $\pm$ 1.67c
0.63	20.33 $\pm$ 1.67b	26.67 $\pm$ 1.67b	35.27 $\pm$ 1.96b
1.25	21.55 $\pm$ 1.65a	35.00 $\pm$ 2.20a	40.00 $\pm$ 2.83a

In a column means with same letter do not significantly differed from each other within varieties at 0.05% level (Tukey's test)

## ANOVA

Source	DF	F value
Concentrations	2	927.11***
Exposure time	2	405.65***
Total	4	

In a column means with same letter do not significantly differed from each other concentration at 0.05% level (Tukey's test). Note: \*\*\*= Significant at  $P < 0.001$ , NS= Non Significant

**Table 6**  $\chi^2$  for heterogeneity, regression equations, LC<sub>50</sub> and 95% confidence limits of different concentrations of spinosad against adult (unsexed) *C. pusillus* after 24, 48 and 72h exposure period

Exposure time (h)	$\chi^2$ for heterogeneity	Regression equation	LC <sub>50</sub> ( $\mu\text{l g}^{-1}$ )	95% confidence limits	
				Lower	Upper
24	0.466	$Y=2.001433+0.9017737X$	12.164	2.089	119.842
48	0.56	$Y=2.215253+0.8069621X$	4.779	1.880	23.671
72	0.122	$Y=3.323893+0.5709572X$	3.654	1.165	25.243

95% confidence limits of LC<sub>50</sub> values, regression equations,  $\chi^2$  for heterogeneity and regression lines of empirical probit mortality of *C. pusillus* are presented in Table 6. The lowest LC<sub>50</sub> value (12.164 $\mu\text{l/g}$ ) was recorded after 72h exhibit that spinosad is toxic against the adult *C. pusillus*. ANOVA revealed high significant effects were found concentrations ( $F = 927.11$ ,  $df = 2$ ,  $P < 0.001$ ) and exposure period ( $F = 405.65$ ,  $df = 2$ ,  $P < 0.001$ ) (Table 5).

#### 4. Repellent activity of Spinosad

The repellent activity test was performed by following the surface film application method. A stock solution was prepared and from this stock solution five doses 1.25, 0.63, 0.32, 0.16 and 0.08  $\mu\text{g/ml}$  were made by serial dilution. One control was used. Half-disc filter papers (Whatman No. 1) were prepared and applied doses of spinosad into half-discs and allowed to dry out in the air. Each treated half-disc then attached lengthwise, edge-to-edge to a control half-disc with a scotch-tape and placed in a petri dish (9 cm diam.). Then 10 adult insects were released in the middle of each filter paper. Insects were counted on the untreated half of the filter paper disc at one hour interval up to five consecutive hours. Three replications were taken and the averages of the counts were converted to percentage repulsion (PR) using the formula of Talukder and Howse (1995).

$PR = (N_c - 5) \times 20$ , where  $N_c$  is the number of insects on the untreated half-disc.

Positive values (+) expressed repellency and negative values (-) for attractant activity.

Repellency of spinosad against *C. pusillus* adults was very much promising, while all the doses found to repel at 0.01% level of significance. Table 7 showed that the F-values were 6.849119 and 6.714021 for the analysis between time interval and doses. In the higher doses the highest repellent activity at 5% ( $P < 0.05$ ) level of significance was observed. The results are in agreement with similar works of Talukder and Howse (1995). Matin and Islam (2016) investigated repellency effect of *Adenantha pavonina* (L.) extracts against pulse beetle, *Callosobruchus chinensis* L. and found all the extracts of *A. pavonina* repellent against *C. chinensis*. Outcome of the experiments indicate that low concentrations spinosad is a potential gustatory toxic agent causing significant mortal effect larva and adult stages of *C. pusillus* at 24-72h exposure. Though, the efficacy of spinosad varied according to the concentrations, the exposure times and as well as the wheat commodities. The larvae impartial of their age became intoxicated by spinosad and so the unsexed adult. The larvae were more capable to spinosad compared to the adults in wheat. Mortality (%) was increased with the concentrations of spinosad and exposure time. The mortal effect was 80.00% mortality in 2<sup>nd</sup> instar larvae at 1.25 $\mu\text{l/g}$  concentration at 72h exposure time.

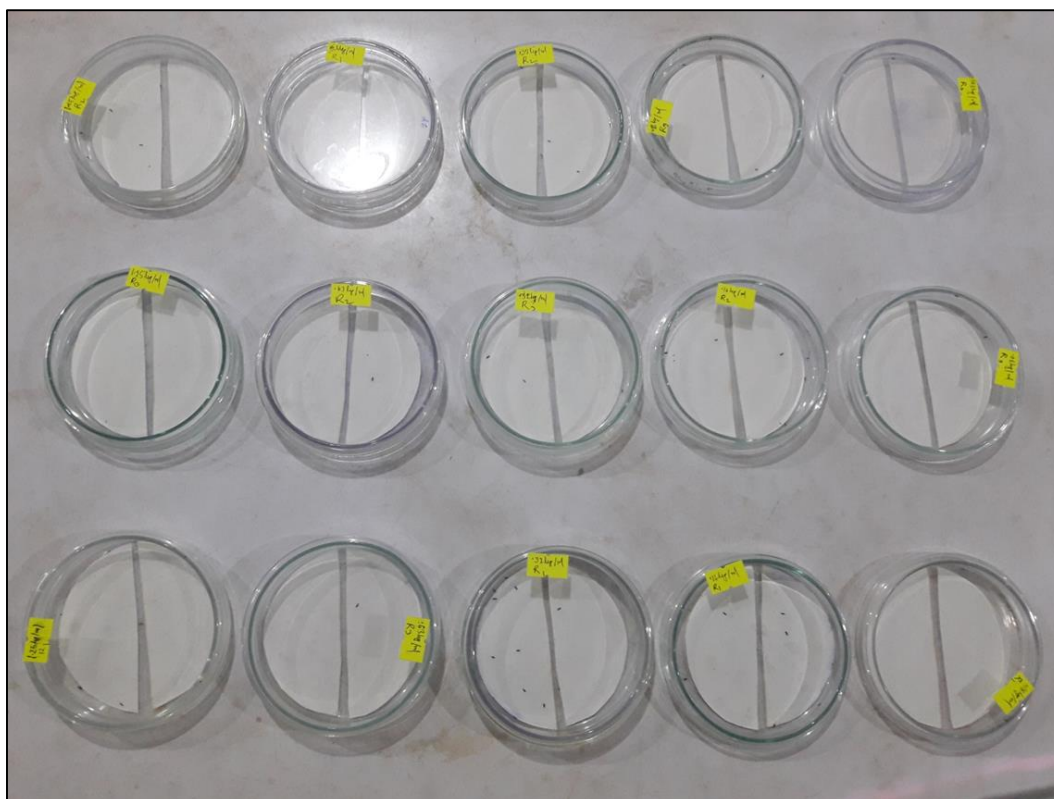
Figured literature showed that larval *C. pusillus* was much doughty to spinosad than their adult (Vayias *et al.* 2010), Fang *et al.* (2002) reported that capability of *Plodia interpunctella* larvae showed dose dependent mortal effect in spinosad treated medium. Towes *et al.* (2003) evaluated that mortality of *C. pusillus* in spinosad treatment and the effect was related to concentrations of soinosad. These results are found to be similar to the present results.

**Table 7** Repellency of *C. pusillus* by spinosed with percent repulsion

Dose	Insect released	Replication	Hour					Average hourly observation (No)					Percentage repulsion PR = (Nc-5) × 20				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
1.25 µg/ml	10	R1	6	8	8	7	6	6.667	7.333	7.333	6.667	6.333	93.34	46.66	46.66	33.34	26.66
		R2	7	8	7	6	6										
		R3	7	6	7	7	7										
0.63 µg/ml	10	R1	8	8	8	8	8	8.333	7.667	7.667	7.667	7.667	66.66	53.34	33.34	53.34	53.34
		R2	8	8	7	8	8										
		R3	8	7	8	7	7										
0.32 µg/ml	10	R1	8	9	8	8	7	8.333	8.00	7.667	7.667	7.667	66.66	60.00	53.34	53.34	53.34
		R2	8	8	7	7	7										
		R3	9	7	8	8	8										
0.16 µg/ml	10	R1	8	8	7	6	6	8.00	7.667	7.333	6.667	6.333	60.00	53.34	46.66	33.34	26.66
		R2	8	8	7	8	7										
		R3	8	7	8	6	6										
0.08 µg/ml	10	R1	8	7	7	7	7	8.00	8.00	7.00	6.667	6.333	60.00	60.00	40.00	33.34	26.66
		R2	9	8	8	7	6										
		R3	7	6	6	6	6										

**ANOVA**

Source of variation	SS	df	MS	F	P-value	F crit
Time (h)	502.4231	4	125.6058	6.849119	0.002066	3.006917
Dose	492.5129	4	123.1282	6.714021	0.00227	3.006917
Error	293.4235	16	18.33897			
Total	1288.359	24				



**Figure 6** Repellency test

Athanassiou *et al.* (2008) found that 3<sup>rd</sup> to 4<sup>th</sup> instar larvae of six *C. pusillus* strains of different locations of Europe were susceptible to spinosad than their adults. In the present results *C. pusillus* adults were found to be less susceptible to spinosad even at higher rate than its young and old larvae. Similar trend of spinosad mortality in wheat were shown by Fang *et al.* (2002), Subramanyam *et al.* (2007). Susceptibility of *C. pusillus* adults to spinosad at the labeled rate (1mg/kg) varied with the wheat class (Sehgal *et al.* 2013).

Toews and Subramanyam (2003) revealed that contact toxicity of spinosad result in 12- 48% mortal effect in adult *C. pusillus*, when exposed to 0.001-0.79mg/cm<sup>2</sup> of spinosad for 24h; and 0, 0.0016 and 0.016mg/cm<sup>2</sup> at 48h exposure. The authors also noted that *C. pusillus* was more tolerant to spinosad than *R. dominica* and *S. oryzae*. Similar results have also been reported by other researchers like Huang *et al.* (2004), Nayak *et al.* (2005), Getchell (2006). Andric *et al.* (2011) reported that *C. pusillus* was less susceptible to spinosad than Nikpay (2007) observed the 65% mortality of *C. pusillus* in treated spinosad at 1mg/kg of wheat; and Athanassiou *et al.*, (2010), observed a maximum 10% adult on wheat and maize.

The larger grain borer, *Prostephanus truncates* and *R. dominica* are very susceptible to spinetoram (product of secondary metabolites spinosyn J and L (Herbert, 2010) to protect grain while *C. pusillus* is less susceptibility to spinetoram (secondary metabolites spinosyn J and L) (Vassilakos and Athanassiou 2012). Spinetoram was effective against *C. pusillus* on wheat (Vassilakos and Athanassiou 2013, Babarinde *et al.*, (2018) noted that a synergistic effect of spinosad with piper guineense and Eugenia aromatic powders causing the death of *C. pusillus* at 3-7 DAT ( days after treatment). Vassilakos *et al.*, (2014) recorded 72.4% mortality against *R. dominica*, Hameed *et al.*, (2012) evaluated the mortality up to 50% against *C. pusillus* for spinosad and two extracts, neem (*Azadirachta indica*) and it can be an effective alternative to synthetic insecticides for eco-friendly management of stored commodity insect pests.

## 5. Conclusion

The present result revealed that spinosad, a reduced- risk bacterial insecticide, can potentially control against the life stages of *C. pusillus* through its contact and gustatory effects. To obtain toxicity concentrations  $\leq 1\mu\text{g}$  of spinosad is enough to produce  $\geq 50\%$  mortality in larval and adult *C. pusillus* in 3 days.



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## Compliance with ethical standards

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### Disclosure of conflict of interest

No conflict of interest.

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