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Contact and Gustatory Effects of Spinosad on the Growth or Survivability and Development of Progeny Buildup in Two Successive Generations of *Cryptolestes pusillus* (Schon.) on Wheat and Rice

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Abstract **Original Research Article**

The contact and gustatory effects of Spinosad on development, survivability, adult emergence and reproductive potential of *Cryptolestes pusillus* (Schon.) in two successive generations were conducted. Egg hatching, larval and pupal survivability, and adult emergence were lowest in wheat and rice in 1st generation, whereas, totally controlled the hatching, larval and pupal survivability and adult emergence of *C. pusillus* was noticed in both the seeds in 2nd generation at 0.63 and 1.25 µg/ml concentrations. The highest PRC value of adult emergence was 97.93% found in wheat and 100% in rice in 2nd generation. The mean developmental period of *C. pusillus* varied in different concentrations of Spinosad. The highest duration of hatching, larval and pupal periods were observed in treated Spinosad at 1.25µg/ml in wheat in F_1 generation, whereas, total control of development was found in F_2 generation in the same concentration.

Keywords: Cryptolestes pusillus (Schon.) Egg hatching, stored grains, environment, Synthetic insecticides.

INTRODUCTION

For the protection of stored grains and other food commodities needs sustainable insect pest management of stored products, considering the safety of the consumers and the environment, within a cost-effective way. *Cryptolestes pusillus* (Schon.) is one of the serious external feeders and common major pests [1], occurring in all areas of the world where grain is produced and store [2, 3]. The damage is caused by both larval and adult stages of *C. pusillus*. Due to high fecundity, polyphagous nature, quick adaptation against insecticides, control of *C. pusillus* for a long time is quite different and rather impossible.

Synthetic insecticides are the currently used pest control method that causes quick mortality of the insects but there are harmful effects of these insecticides on the health [4], environment [5], and on non-targeted organisms [6]. Stored grain insects have developed resistance to these insecticides [7]. As the treated grains are consumed by human, there is a need to use reduced risk insecticides as an alternate to conventional insecticides [8].

Spinosad is an insecticide product from Dow AgroSciences (Indianapolis, Indiana, USA), derived via fermentation from a naturally-occurring soil actinomycete, *Saccharopolyspora spinosa* Mertz and Yao (Bacteria: Actinobacteria). It is registered in several countries as a grain protectant at the maximum labelled use rate of 1mg/Kg of grain and its Maximum Residue Limit (MRL) established at 1.5ppm [9]. The Spinosad can persist from 6-12 months on the stored commodities [10-14].

So far, the effect of Spinosad on *C. pusillus* have been conducted in a very few levels in Bangladesh. The aim of this study was to evaluate the effect of Spinosad on the survivability and development of progeny buildup in two successive generations of *C. pusillus* under laboratory conditions.

MATERIALS AND METHODS

One gram of wheat and rice were socked in different concentrations of Spinosad separately and then dried at room temperature for 24h in a 6cm glass petri dish. Deposited eggs 24-h old of *C. pusillus* were collected by sieving the culture food medium maintained in the past five years in Control Temperature (CT) room and released on the treated wheat. After introduction the eggs, the petri-dish was covered and kept until the eggs began to hatch. It was checked for hatching up to 10 days. Newly hatched larvae were collected carefully with fine camel hair brush after 4-5 days, and then transferred to wheat medium treated with different concentrations of Spinosad. The larvae were reared up to the adult emergence. After every three days the wheat was changed with uninfected wheat. A similar set of experiment was carried on wheat soaked with distilled water only, as a control batch. The temperature was maintained at 30±1^{0}C with 75% RH in the CT room throughout the study periods.

Egg-to-larval survival of *C. pusillus* was checked after 21 days, while egg-to-adult emergence, progeny production for this species recorded 42 days. Adults unable to move when prodded gently with a hair brush were considered dead. Each combination of *C. pusillus*, Spinosad concentrations, and exposure duration was replicated three times, and each replicate was treated separately in wheat and rice. Progeny production or egg-to-adult emergence and developmental period of life stages of *C. pusillus* was counted to untreated wheat and treated wheat with Spinosad concentration for two successive generation.

Parameters observed:

Effect of spinosad was observed on the following biological parameters of *C. pusillus* in two successive generations:

A. Growth or Survivability of life stages of *C. pusillus* on wheat-

i. Hatching percentage,
ii. larval survivability up to pupation,
iii. pupal survivability and
iv. adult recovery or emergence

B. Developmental period of life stages of *C. pusillus* on wheat.

i. Hatching period,
ii. larval period and
iii. pupal period

Statistical analysis:

All data were subjected to Analysis of variance using SPSS-20 version. Means comparisons were performed by Tukey's tests (P<0.05) using MS excel-2010. PRC was calculated [15].

RESULTS AND DISCUSSION

i. Effect on hatching

Hatching of *C. pusillus* in different concentrations of Spinosad was significantly (P<0.001) lower in treated wheat compared with untreated wheat in a dose-dependent manner in both F_1 and F_2 generations (Table 1). In higher concentrations (1.25µg/ml) 47.61±3.38/d was observed but in lower concentrations (0.08 µg/ml) was recorded (77.15±3.52%). The number of hatched larvae was decreased with the increase of Spinosad concentrations in F_1 generation. In F_2 generation, only (5.20± 1.70% egg hatching was observed in higher concentrations in treated wheat. In treated wheat the egg hatchability was ranged from 47.61± 3.38 to 77.15±3.52% in F_1 generation but it was 4.20±1.79 to 44.78±2.88% in F_2 generation. But it was 88.12±2.02% in F_1 generation and 80.76±4.87% in F_2 generation in control. Spinosad highly influenced the egg hatchability of *C. pusillus* in wheat in different concentrations (F=24.67, df+5, P<0.001) in F_1 and (F=78.92, df=5, P<0.001) in F_2 Generations.

In rice treated seeds, 18.46±1.67% to 44.33±1.12% of hatchability was recorded in F_1 generation but no hatchability was recorded in higher concentrations in F_2 generation (Table 3). Significant differences were noted in F_1 (F=73.22, df=5, P<0.001) and (F=66.95, df=5, P<0.001) in F_2 generation.

ii) Larval survivability:

The different concentrations of Spinosad significantly influenced the larval survivability of *C. pusillus* in treated wheat and rice respectively. The highest larval survivability was recorded at 1.25µg/ml of Spinosad was 47.61±3.35% but lowest 77.15±3.52% in wheat treated seeds in F_1 generation whereas only 2.00±0.80% in higher concentrations but 30.14±3.21% in lower concentration in F_2 generations (Table1-2). Significant effect of concentration of Spinosad was noticed in F_1 generation (F=19.21, df=5, P<0.001) and F=78.92, df=5, P<0.001) in F_2 generation.

On the other hand, 22.22V1.87% larval survivability was observed in higher concentration but 76.16±3.03% in lower concentration in rice treated seeds in F_1 generation but no larval survivability was recorded in F_2 generation. Significant result was observed (Table 3 & 4).

iii) Pupal survivability:

Spinosad treatment significantly reduced pupal survivability in *C. pusillus* in wheat treated seeds in both F_1 and F_2 generations. The range of pupal survivability was 50.86±3.08 to 82.10±2.88 in higher and lower concentrations in F_1 generation in wheat treated seeds, but in F_2 generation no pupal survivability was noticed in higher concentrations. Significant results were obtained in F_1 (F=25.07, df=4, P<0.001) generation but in F_2 it was (F=159.64, df=4, P<0.001).

The range of pupal survivability was recorded 36.26±4.06 to 74.25±3.115 in higher concentration in F_1 generation in rice treated seeds. No pupal survivability was recorded in F_2 generation like in wheat. Significant pupal survivability was noticed in F_2 generation.

iv) Adult Emergence:

Tables (1-4) shows that the percentage of adult emergence in both F_1 and F_2 generations of wheat and rice treated seeds of Spinosad. It was 52.24±3.98 to 90.60±4.22% in higher and lower concentrations in wheat treated seeds in F_1 generation. Significant was obtained. But in rice, 32. ±26 to 77.60±3.67% of adult recovery was resulted in F_1 generation. No adult recovery was obtained in F_2 generation of rice treated seeds.

B. Development periods

i) **Effect on hatching period**: All concentrations of Spinosad was influenced of hatching period compared with control, in a dose-dependent manner (Tables 3-4). Both F_1 and F_2 generations, hatching period was longest significant in both wheat and rice seeds. Lower hatching was recorded in lower concentrations but higher in higher concentrations (1.25 µg/ml). It was 5.00±0.41 to 8.90±0.49 in F_1 but 6.16±0.45 to 12.80±0.57 in F_2 generations. The Statistical analysis revealed that significant differences were present among the concentrations (F=26.33, df=4, P<0.001) in F_1 and (F=26.-6, df=4, P<0.001 in wheat but 113.01, df=4, P<0.001 in rice) in F_2 generations respectively.

ii) **Effect on larval period**: Spinosad gradually increased in higher concentrations in both wheat and rice seeds. The larval duration was ranged from 25.25±12.23 to 30.00±2.36 in wheat and 21.52±0,59 to 28.50±0.55 in rice of F_1 generation whereas no larval duration was observed in higher concentrations of F_2 generations in wheat and rice. Analysis of variance showed significant differences was noticed in F_2 generations in both wheat and rice.

iii) **Effect on pupal period**: The pupal period was shortest in lower concentration but highest in higher concentrations (Table 3) both in F_1 and F_2 generations in wheat and rice respectively. There is no larval development occurred in higher concentrations in F_2 generation in both wheat and rice. The longest pupal period 12.08±0.46 and 12.12±0.41 in rice. Significant difference in pupal period were noticed.

Table 1: Effect of spinosad on Growth or survivability at different stages and adult emergence of *C. pusillus*in F_1 generation on wheat

Concentration µg/ml		F_1 **Generation in wheat**			
	Hatching Survivality	**Larval Survivality**	**Pupal Survivality**	**Adult Survivality**	**PRC**
Control	88.12±2.02a (88.00-90.0)	80.24±2.47a (80.00-81.00)	88.24±2.66a (88.00-89.00)	92.52±2.50a (92.00-94.00)	-
0.08	77.15±3.52ab (77.00-80.00)	80.24±2.94a (80.00-82.00)	82.10±2.88a (82.00-83.00)	90.60±4.22a (90.00-92.00)	5.45
0.16	80.06±3.00ab (80.00-81.00)	78.20±3.04a (78.00-79.00)	81.06±2.17a (81.00-82.00)	88.80±2.16a (88.00-90.00)	6.02
0.32	72.00±3.64bc (72.00-74.00)	72.10±3.73ab (72.00-74.00)	78.02±3.34a (78.00-80.00)	78.60±4.43a (78.00-80.00)	13.86
0.63	60.12±1.54cd (60.00-61.00)	62.12±3.21b (62.00-65.00)	63.05±2.59b (63.00-65.00)	61.50±2.29b (61.00-63.00)	29.31
1.25	47.61±3.38d (47.00-50.00)	45.42±3.31c (45.00-47.00)	50.86±3.08b (50.00-52.00)	52.24±3.98b (52.00-54.00)	43.82

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

Table 2: Effect of spinosad on Growth or survivability at different stages and adult emergence of *C. pusillus*in F_2 generation on wheat

Concentration µg/ml	F_2 **Generation on wheat**				
	Hatching Survivality	**Larval Survivality**	**Pupal Survivality**	**Adult Survivality**	**PRC**
Control	80.76±4.87a (80.00-82.00)	60.26±3.12a (60.00-61.00)	78.42±2.43a (78.00-80.00)	80.00±1.16a (80.00-81.00)	-
0.08	44.78±2.88b (44.00-45.00)	30.14±3.21b (30.00-33.00)	50.40±2.70b (50.00-52.00)	44.44±2.38b (44.00-46.00)	43.31
0.16	28.68±1.30c (28.00-29.00)	26.26±4.47b (26.00-28.00)	28.06±2.54c (28.00-30.00)	10.43±2.72c (10.00-12.00)	68.80
0.32	20.40±4.81cd (20.00-23.00)	20.08±3.45bc (20.00-23.00)	16.00±3.06d (16.00-18.00)	4.05±2.98cd (4.00-6.00)	79.79
0.63	8.24±1.15de (8.00-9.00)	7.05±1.51cd (7.00-8.00)	6.00±2.08de (6.00-7.50)	0.00±0.00d (00.0-00.0)	92.89

1.25	4.20±1.70e (4.00-5.00)	2.00±0.80d (2.00-2.50)	0.00±0.00e (00.0-00.0)	0.00±0.00d (00.0-00.0)	97.93

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

Table 3: Effect of spinosad on Growth or survivability at different stages and adult emergence of *C. pusillus*in F_1 generation on Rice

Concentration µg/ml	F_1 Generation on Rice				
	Hatching Survivality	**Larval Survivality**	**Pupal Survivality**	**Adult Survivality**	**PRC**
Control	72.00±2.53a (72.00-74.00)	70.00±1.08a (7.00-8.00)	78.12±4.14a (78.00-80.00)	80.12±2.01a (80.00-82.00)	-
0.08	44.33±1.12b (44.33-45.00)	76.16±3.03a (76.00-78.00)	74.25±3.11a (74.00-77.00)	77.60±3.67ab (77.00-79.00)	9.48
0.16	36.26±3.16bc (36.00-37.00)	70.08±3.54a (70.00-72.00)	70.20±1.56a (70.00-71.00)	60.24±4.50bc (60.00-63.00)	21.89
0.32	32.12±3.12c (32.00-35.00)	64.34±3.93ab (64.00-67.00)	66.02±2.51a (66.00-68.00)	52.12±2.76cd (52.00-54.00)	31.98
0.63	20.14±1.21d (20.00-21.00)	52.12±3.90b (52.00-55.00)	44.05±4.98b (44.00-47.00)	40.24±4.36de (40.00-43.00)	48.82
1.25	18.46±1.67d (18.00-19.00)	22.22±1.87c (22.00-23.00)	36.26±4.06b (36.00-39.00)	32.26±5.19e (32.00-34.00)	64.20

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

Table 4: Effect of spinosad on Growth or survivability at different stages and adult emergence of *C. pusillus*in F_2 generation on Rice

Concentration µg/ml	F_2 Generation on Rice				
	Hatching Survivality	**Larval Survivality**	**Pupal Survivality**	**Adult Survivality**	**PRC**
Control	72.12±3.58a (72.00-74.00)	66.56±3.45a (66.00-67.00)	70.72±1.30a (70.00-71.00)	68.24±4.33a (68.00-70.00)	-
0.08	50.00±4.30b (5.00-6.00)	44.28±6.81b (44.00-46.00)	40.24±4.05b (40.00-42.00)	30. 5 21±.15b (30.00-31.00)	47.69
0.16	24.00±4.73c (24.00-26.00)	20.10±2.50c (20.00-21.00)	18.20±4.31c (18.00-20.00)	30.20±5.14b (30.00-32.00)	70.56
0.32	18.00±4.48c (18.00-20.00)	4.40±2.34cd (4.00-5.00)	2.24±1.43d (2.00-3.00)	0.00±0.00c (00.0-00.0)	90.89
0.63	0.00±0.00d (00.0-00.0)	0.00±0.00d (00.0-00.0)	0.00±0.00d (00.0-00.0)	0.00±0.00c (00.0-00.0)	100.00
1.25	0.00±0.00d (00.0-00.0)	0.00±0.00d (00.0-00.0)	0.00±0.00d (00.0-00.0)	0.00±0.00c (00.0-00.0)	100.00

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

Results of the present experiments revealed that there were significant impacts of the survivability and developmental period of *C. pusillus* in different concentrations of Spinosad and exposure periods. It was noted Spinosad concentrations from 0.08 to 1/25µg/ml were found to be highly effective against *C. pusillus* by decreasing the survivability of life stages of *C. pusillus* egg hatching duration, and length of larval and pupal period in F_1 and F_2 generation than control. It completely controlled the larval and pupal survivability (0.00±0.00%) and adult emergence (0.00±0.00%) in wheat and rice seeds at 0.63 and 1.25µg/ml concentration in F_2 generation. There was significant effect in egg hatching to adult emergence percentage and egg hatching period, larval and pupal period in wheat and rice seeds in F_1 and F_2 generation. Similar studies mentioned the pesticidal potentials of Spinosad against a few stored product insect pest [16, 17]. The present results of Spinosad are well accordance with the results of the mentioned reports.

Spinosad at high concentration (1.25µg/ml) able to control the adult *C. pusillus*. It was affected egg to larval survival of *C. pusillus*. Reduction in larval and pupal survival was observed in wheat and rice at all concentrations. Mollaie *et al*., [18] evaluated that 0.1-1mg/kg of spinosad absolutely suppressed the larval survival of *E. kuehniella* and also noted that 1mg/kg of Spinosad suppressed larval survivability and adult emergence of *P. interpunctella* greater than 90%; and this result are more or less accordance with the present result.

Fang *et al*., [19], Huang *et al*., [20] and Huang and Subramanyam [21] indicated that susceptibility of *P. interpuntella* larvae to Spinosad was dose dependent. Larval mortality on Spinosad approached 88% at 22 C temperature.

The present results indicate that Spinosad is an effective tool to control all the life stages of *C. pusillus*. The degree of toxicity of Spinosad can be rankled as concluded that low concentration of Spinosad would be potential to control *C. pusillus* in storage system. It is a very safe method for food preservation and pest control.

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