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Toxicity evaluation of Spinosad against Malathion and deltamethrin resistant population of rusty grain beetle, *Cryptolestes ferrugenius* (Stephens) in Andhra Pradesh, India

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Abstract
A laboratory experiment was implemented in the Department of Entomology Agricultural College, Bapatla to know the resistance levels of malathion and deltamethrin against Rusty grain beetle, *Cryptolestes ferrugenius* (Stephens) and management with novel insecticide molecule Spinosad. The degree of resistance developed by resistant population of *C. ferrugenius* shows 34.66 and 21.35 folds resistant at LC_{50} level; 4.14 and 9.89 folds resistant at $LC_{99.9}$ level to malathion and deltamethrin, respectively in comparison with laboratory susceptible strain at 72 HAT. Insecticide tested for the management of malathion and deltamethrin resistant population of *C. ferrugenius*, Spinosad was found to be the finest at $LC_{99.9}$ level with the relative toxicity (3.43 and 1.48 times) than malathion and deltamethrin respectively.

Keywords: *Cryptolestes ferrugenius,* Malathion, Deltamethrin, Spinosad, Resistance

Introduction
Annually storage losses were estimated as 14.02 million tons of food grains worth of Rs. 7145 crores of every year in India. Food grains constitute the bulk of diet of the people. In India, with the advent of green revolution, the food grain production has gone up from 50 M to 291.95 M t in 2020 [1]. In India large quantities of food grains are stored by Food Corporation of India, Central and State Warehousing Corporations and other agencies not only to maintain adequate food supply but also to meet buffer and reserve stocks to face the natural calamities. Total food grain losses expected due to insect pests account for nearly Rs.1275 crores [2]. In India, approximately 10 percent of food grains were lost during storage and half of which is due to insect pests [3]. Accountable losses due to insect pests are 3.4 per cent, rodents 0.4 to 1.0 percent, moisture 0.2 percent and other sources 0.3 percent [4].
Among the several storage insect pests, rusty grain beetle *C. ferrugineus* (Stephens) (Cucujidae: Coleoptera), is an important and destructive insect pests of stored cereals, oilseeds, miscellaneous food grains and is cosmopolitan in distribution. It feeds on the germ portion of the grain in preference to endosperm [5]. Over the past four decades, there has been a sustained research activity in this storage pest. Development of insecticide resistance in insect pests of crops has become a major constraint both in crop protection and crop production. The occurrence of insecticide resistance was first esteemed in Sanjose scale, *Quadraspidiotus periniciosus* (Comstock) [6]. Due to the resistance by insect pests and negative effects of synthetic pesticides to the environment, it is the perfect time to use novel and suitable compounds in insect pest's management.

Materials and Methods
Rearing of the test insect
The adult beetles of *C. ferrugineus* collected from the storage house of Andhra Pradesh were preserved on wheat grains in laboratory during 2018. About 200 g of the flour was taken in a plastic jar measuring 45 × 15 cm and about 50 adults were released into it for oviposition. The test insect populations were managed at an optimum temperature of 32 ± 1 ˚C and 75± 2 per cent relative humidity all over the period of investigation (Andrewartha, 1961). After seven days, the adults were removed and released into another jar containing the diet,

Thus a succession of jars were maintained for utilizing the eggs laid staggeredly to ensure a constant supply of test insects of known age. After hatching of eggs, the diet in the jars was changed once in a week by sieving and transferring the pest stages to fresh jars containing diet upto pre-pupal stages. The newly emerged adults were shifted into a fresh diet and used for the multiplication of culture as well as for bioassay studies.

Test insect population

The adult beetles of C. ferrugineus (One days old) of F1 generation, reared in the laboratory from the initial culture obtained from the places *viz.*, Bapatla, Machilipatnam, Rajahmundry were subjected to bioassay. The laboratory culture of C. ferrugineus which is being maintained in the Department of Entomology Agricultural College, Bapatla, without any insecticidal exposure and were sustained as susceptible strain.

Bioassay by Jute cloth disc impregnation method

The adult beetles of C. ferrugineus of one day old were subjected to the bioassay with the test insecticides by following jute cloth disc impregnation method. One per cent concentration of stock solution of test insecticides was prepared by weighing the required quantity of malathion and deltamethrin and dissolved in acetone. The graded concentrations of the test insecticides were prepared with acetone as solvent by following serial dilution technique. Two ml of insecticidal solution was found to be sufficient for complete impregnation of the jute cloth disc of nine cm diameter. After impregnation, the jute cloth discs were air dried. One week old beetles were collected from the culture and were kept under starvation for two hours. The starved beetles were transferred to the petri plates containing insecticide impregnated jute cloth disc @ 20 beetles/ petri plate in three replications for each test insecticide. The insects were confined to the treated surface for 24 hours and survivals were transferred into a fresh petriplate with diet. Simultaneously, a control was also maintained which containing jute cloth disc impregnated with acetone only.

Data Collection

Mortality data were recorded at 24, 48 and 72 hours after treatment (HAT). A preliminary experiment was conducted with wide range of concentrations followed by a narrow range to get mortality in the range of 5-90%. There was no occasion of the use of Abbott's formula since mortality was not recorded in control, because of the use of almost same age beetles and the experiment conducted under controlled ambient conditions of 32 ˚C temperature and 75 per cent relative humidity. Moreover, a separate set of petri plates were utilized for control avoiding the use of petri plates that were used for insecticidal treatments. In order to know the immediate toxicity of the chemical, the mortality data were recorded at 24 HAT and subsequently at 48 HAT. Mortality data at 72 HAT was also recorded to know the mortality end point.

Assessment of the Degree of Resistance

The mortality data of adult beetles of resistant machilipatnam strains against all the test insecticides were subjected to probit analysis [7] using SPSS 16.0 (Statistical Package for Social Sciences) software to calculate LC50, LC99.9, heterogeneity ($\chi2$), intercept: (a), slope of the regression line (b), regression equation and fiducial limits. The degree of resistance acquired by C. ferrugineus was calculated for the data at 72 HAT by dividing the higher LC50 / LC99.9 value of each strain with the least LC50 / LC99.9 value and thus the relative degree of resistance was assessed.

Results and Discussion

Resistance of malathion and deltamethrin to C. ferrugineus

The resistant population of C. ferrugineus recorded LC50 and LC99.9 values as 0.225 and 6.35 per cent, respectively to malathion at 24 HAT. The corresponding values slightly decreased with increased mortality at 48 HAT (0.169 % and 5.11 %) and at 72 HAT (0.104 % and 4.90 %). The slope (b) values of log concentration probit (lcp) lines of malathion were 1.87, 1.78 and 1.87 at 24, 48 and 72 HAT, respectively. The chi square test revealed the homogeneity of the test population ($P<0.05$) (Table 1). The degree of resistant acquired by resistant population of C. ferrugineus 34.66 folds resistant at LC50 and 4.14 folds resistant at LC99.9 level to malathion in comparison with laboratory susceptible strain at 72 HAT (LC50 = 0.003 % and LC99.9 = 1.184 %) (Table 3).

The data revealed that LC50 and LC99.9 values of deltamethrin with resistant strain of C. ferrugineus at 24 HAT were 0.244 and 6.519 per cent, respectively. At 48 and 72 HAT there was a slight increase in the mortality with consequent decrease in the LC50 (0.181and 0.085 %) and LC99.9 (4.558 % and 2.120 %) values, respectively. The slope (b) values of log concentration probit (lcp) lines were 3.32, 3.10 and 3.00 at 24, 48 and 72 HAT, respectively. The chi square test revealed the homogeneity of the test population ($P<0.05$) (Table 2).

Table 1: Relative toxicity of malathion to resistant and susceptible population of rusty grain beetle, *C. ferrugenius*

HAT	LC_{50} (95% FL) (%)	$LC_{99.9}$ (95% FL) (%)	Slope b (±SE)	Heterogeneity (χ^2)	Regression equation (Y=a+bx)
Resistant population					
24	0.225 (0.164-.449)	6.35 (3.45-24.225)	1.87 (±0.201)	16.13	Y=1.58+1.87x
48	0.169 (0.124-.291)	5.11 (2.50-20.121)	1.78 (±0.165)	17.92	Y=2.57+1.78x
72	0.104 (0.062-.197)	4.90 (2.09-13.121)	1.87 (±0.133)	21.73	Y=3.17+1.87x
Susceptible population					
24	0.011 (0.008-0.015)	1.987 (0.653-6.121)	2.22 (±0.119)	4.66	Y=1.12+2.22x
48	0.006 (0.004-0.010)	1.671 (0.451-4.375)	1.56 (±.109)	2.02	Y=1.51+1.56x
72	0.003 (0.049-6.470)	1.184 (0.049--2.421)	1.43 (±.106)	5.54	Y=1.86+1.43x

Table 2: Relative toxicity of malathion to resistant and susceptible population of rusty grain beetle, *C. ferrugenius*

HAT	LC_{50} (95% FL) (%)	$LC_{99.9}$ (95% FL) (%)	Slope b (±SE)	Heterogeneity (χ^2)	Regression equation (Y=a+bx)
Resistant population					
24	0.244(0.018-0.468)	6.519(0.365-8.063)	3.32(±0.291)	3.15	Y=2.00+3.32x
48	0.181(0.007-0.210)	4.558(0.121-6.844)	3.10(±0.228)	4.47	Y=2.50+3.10x
72	0.0854(0.067-0.153)	2.120(0.343-4.188)	3.00(±0.214)	5.05	Y=3.80+3.0x
Susceptible population					
24	0.008(0.006-0.015)	0.978(0.323-3.647)	2.57(±0.175)	10.007	Y=1.20+2.57x
48	0.006(0.005-0.009}	0.877(0.251-2.492)	2.50(±0.190)	7.906	Y=2.00+2.50x
72	0.004(0.003-0.005)	0.659(0.128-1.247}	2.41(±0.225)	12.451	Y=2.07+2.41x

*Values in parenthesis indicates Fiducial Limits (FL)
HAT- Hours after treatment

Table 3: Relative toxicity of malathion to resistant and susceptible population of rusty grain beetle, *C. ferrugenius*

HAT	LC_{50} (95% FL) (%)	$LC_{99.9}$ (95% FL) (%)	Slope b (±SE)	Heterogeneity (χ^2)	Regression equation (Y=a+bx)
Resistant population					
24	0.225 (0.164-.449)	6.35 (3.45-24.225)	1.87 (±0.201)	16.13	Y=1.58+1.87x
48	0.169 (0.124-.291)	5.11 (2.50-20.121)	1.78 (±0.165)	17.92	Y=2.57+1.78x
72	0.104 (0.062-.197)	4.90 (2.09-13.121)	1.87 (±0.133)	21.73	Y=3.17+1.87x
Susceptible population					
24	0.011 (0.008-0.015)	1.987 (0.653-6.121)	2.22 (±0.119)	4.66	Y=1.12+2.22x
48	0.006 (0.004-0.010)	1.671 (0.451-4.375)	1.56 (±.109)	2.02	Y=1.51+1.56x
72	0.003 (0.049-6.470)	1.184 (0.049--2.421)	1.43 (±.106)	5.54	Y=1.86+1.43x

Table 4: Relative toxicity of malathion to resistant and susceptible population of rusty grain beetle, *C. ferrugenius*

HAT	LC_{50} (95% FL) (%)	$LC_{99.9}$ (95% FL) (%)	Slope b (±SE)	Heterogeneity (χ^2)	Regression equation (Y=a+bx)
Resistant population					
24	0.244(0.018-0.468)	6.519(0.365-8.063)	3.32(±0.291)	3.15	Y=2.00+3.32x
48	0.181(0.007-0.210)	4.558(0.121-6.844)	3.10(±0.228)	4.47	Y=2.50+3.10x
72	0.0854(0.067-0.153)	2.120(0.343-4.188)	3.00(±0.214)	5.05	Y=3.80+3.0x
Susceptible population					
24	0.008(0.006-0.015)	0.978(0.323-3.647)	2.57(±0.175)	10.007	Y=1.20+2.57x
48	0.006(0.005-0.009}	0.877(0.251-2.492)	2.50(±0.190)	7.906	Y=2.00+2.50x
72	0.004(0.003-0.005)	0.659(0.128-1.247}	2.41(±0.225)	12.451	Y=2.07+2.41x

*Values in parenthesis indicates Fiducial Limits (FL)
HAT- Hours after treatment

Table 5: Relative toxicity of newer insecticide Spinosad in comparison with Malathion and Deltamethrin against resistant population of rusty grain beetle, *C. ferrugenius* for its management at 72 HAT

Hours after treatment	Relative toxicity of Spinosad (45 % EC)			
	Over Malathion		Over Deltamethrin	
	LC_{50}	$LC_{99.9}$	LC_{50}	$LC_{99.9}$
24	2.79	1.47	3.03	1.51
48	3.14	2.34	3.37	2.09
72	4.14	3.43	3.40	1.48

The Machilipatnam showed 21.35 folds resistance at LC50 and 9.89 folds resistance at LC99.9 level to deltamethrin in comparison with Laboratory susceptible strain (LC50 = 0.004 % and LC99.9 = 0.659 %) at 72 HAT (Table 3). In the current study the LC50 and LC99.9 values of malathion for Machilipatnam were 0.104 and 4.90 % which were lower than the values (0.17 and 8.4793) reported by [8] which revealed decrease in the levels of resistance to malathion which may be decrease in usage of malathion and due to the alternative usage of deltamethrin in the godowns. Different studies in the past have also shown that many organo-phosphate insecticides as well as malathion resulted in the development of resistance in many insect species including lesser grain borer [9, 10].

Sublethal doses of malathion greatly suppressed acid phosphatase activity in resistant beetles, which might be due to inhibition of this enzyme under insecticidal toxicity and impairing of the lysosomal activity to hydrolyze the macromolecules and in turn limiting the ability of the resistant beetles to use energy rich compounds to obtain energy. The present results were on par with those of [11], who reported that a strain of *T. granarium* has developed resistance which is ranged from 1.55 to 3.65 folds in six successive generations at 80 percent selection pressure.

Efficacy of Spinosad against malathion and deltamethrin resistant population

It was estimated from the data (Table 4) that the LC_{50} values of spinosad to the adults of resistant strain of *C. ferrugineus* were 0.0804, 0.0537 and 0.0251 per cent while the respective $LC_{99.9}$ values were 4.3036, 2.1751 and 1.4254 at 24, 48 and 72 HAT, respectively. The slope (b) values of log concentration probit (lcp) lines for spinosad were 1.20, 1.14 and 1.11 at 24, 48 and 72 HAT, respectively. The chi-square test revealed that the population used in the study was homogenous (P<0.05).

Spinosad LC_{50} level was 2.79, 3.14 and 4.14 times more toxic than malathion at 24, 48 and 72 HAT, respectively. The corresponding value was 3.03, 3.37 and 3.40 times more toxic than deltamethrin at 24, 48 and 72 HAT. At $LC_{99.9}$ level less relative toxicity in comparison with malathion and deltamethrin (Table 5).

The LC_{50} and $LC_{99.9}$ values insecticide was spinosad (0.0251 % and 1.4254 %) against *C. ferrugineus*. In present study spinosad was relatively less toxic as already reported by [12, 13] due to less usage in godowns. However, usage of spinosad at high rates may be cost-prohibitive. Similarly, [14] also reported complete control of *R. dominica* adults when exposed to a dust formulation of spinosad (0.125 a.i. D) on corn treated with 0.7 to 1.4 mg.kg-1 seed. Similarly [15] recorded Laboratory and field data have shown spinosad at 1 mg ([AI])/kg to be effective against several major stored-grain insects including *C. ferrugineus.* Similarly, a residual deposit of spinosad @ 0.05 and 0.1 mg.cm-2 of surfaces *viz*., concrete surface, galvanized floor, un waxed floor tile and waxed floor tile gave cent per cent mortality of adults of eight stored product beetles *viz., C. ferrugineus, R. dominica, T. castaneum, T. confusum, T. variabile, S. oryzae and O. surinamensis* [16].

Conclusion

From the present study we have seen that spinosad 45 % EC was found to be effective with high relative toxicity (4.14 and 3.40 times) than malathion 50 % EC and deltamethrin 2.8 % EC at LC_{50} level for the management of resistant Machilipatnam population of *C. ferrugineus*. It can be concluded from this study that resistant Machilipatnam populations of *C. ferrugineus* have developed resistance against organo-phosphate insecticides but it can be partially controlled by spinosad. Higher doses or other pesticides are required to control resistant population. Secondly, development of resistance and the susceptibility in insects is a biochemical phenomenon which is evident from number of macromolecular derangements resistant and susceptible populations of *C. ferrugineus*. Future direction of the study to demonstrates the importance of research to develop newer insecticides and innovative strategies to prevent the development of insecticide resistance.

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References

1. Agricultural statistics at a glance, Ministry of Agriculture (Department of Agriculture and Cooperation, Directorate of Economics & Statistics), 2019-20.
2. Mohan S, Kavitharaghavan Z. Studies on the popularization of TNAU stored product insect management kit technology. Green Farming, 2008, 46-53.
3. Narang DD. Banish the Pests Feeding on our Grain. In the Tribune September 23, Chandigarh, 2002, 168-170.
4. Srinivasa V, Reddy, KD and Sridhar Y. Management of *Sitophilus zeamais* in stored maize using grain protectants. Indian Journal of Entomology. 2003; 65(4):492-495.
5. Pingale SV. Insects and mites attacking food grain in storage. Indian Council of Agricultural Research, New Delhi, 1976, 186-188.
6. Melander AL. Can insects become resistant to sprays? Journal of Economic Entomology. 1914; 7:167.
7. Finney DJ. Probit analysis. Cambridge University Press, London, 1971, 109-110.
8. Madhumathi T, Subbaratnam GV, Murthy MLN, Jayaraj S. Insecticide resistance in *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Cucujidae) in Andhra Pradesh. Pest Management and Economic Zoology. 2000; 8(1):5-10.
9. Badmin JS. IRAC survey of resistance of stored grain pests: Results and Progress. Proceedings of the Fifth International Working Conference on Stored Product Protection. Bordeaux, France, 1990, 973-982.
10. Rossiter LC, Gunning RV, Rose HA. The use of Polyacrylamide Gel Electrophoresis for the investigation and detection of fenitrothion and chlorpyrifos-methyl resistance in *Oryzaephilus surinamensis* (L.) (Coleoptera, Silvanidae). Pesticide Biochemistry and Physiology. 2001; 69(1):27-34.
11. Kumar MK, Srivastava C, Garg AK. *In vitro* selection of deltamethrin resistant strain of *Trogoderma granarium* and its susceptibility to insecticides. Annals of Plant Protection Sciences. 2010; 8(1):26-30.
12. Subramanyam B, Fang L, Arthur FH. Effectiveness of spinosad on four classes of wheat against five stored product insects. Journal of Economic Entomology. 2002; 95(3):640-650
13. Daglish GJ, Nayak MK, Byrne VS. Is there a role for spinosad in protecting Australian grain from insect. Proceedings of the Australian Post Harvest Technical Conference, 2003, 78-81.
14. Mutambuki KCM, Ngatia J, Mbugua N, Likhayo P. Evaluation of the efficacy of spinosad dust against major stored insect pests. Proceedings of 8th International Working Conference on Stored Product Protection, July 22-26, Newyork, UK, 2002, 888-891.
15. Toews MD, Subramanyam B. Contribution of contact toxicity and wheat contribution to mortality of stored product insects exposed to spinosad. Pest Management Science. 2003; 59:538-544.
16. Toews MD, Subramanyam B, Rowan JM. Knockdown and mortality of adults of eight species of stored - product beetles exposed to four surfaces treated with spinosad. Journal of Economic Entomology. 2003; 96(6):1967-1973.