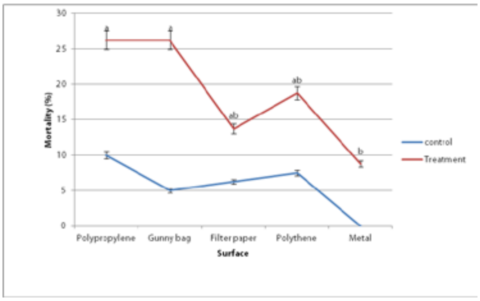

Figure 1. Mortality of *Tribolium castaneum* adults exposed to spinosad-treated surfaces for 2 days.

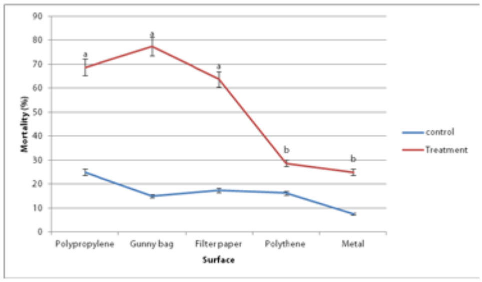

Figure 2. Mortality of *Tribolium castaneum* adults exposed to spinosad-treated surfaces for 6 days.

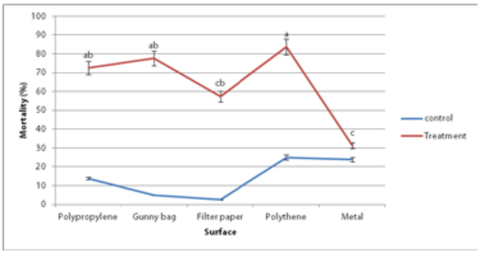

Figure 3. Mortality of *Rhyzopertha dominica* adults exposed to spinosad-treated surfaces for 2 days.

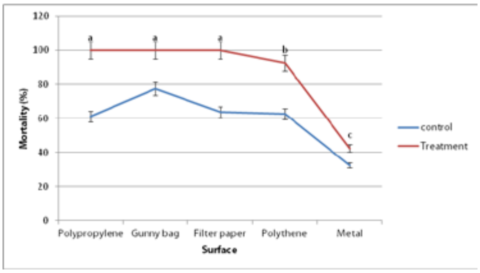

Figure 4. Mortality of *Rhyzopertha dominica* adults exposed to spinosad-treated surfaces for 6 days.

References

ARTHUR, F.H, 2007. Insect pest management in stored products using reduced-risk insecticides. In: Navarro, S., Adler, C., Riudavets, J., Stejskal, V. (eds.), Proceedings of the IOBC/WPRS Working Group "Integrated Protection of Stored Products", September 20-23, 2005, Prague, Czech Republic, Institute for Biological Control, Darmstadt, 233-241.

BOINA, D.R., SUBRAMANYAM, B., MUTAMBUKI, K. 2012. Delayed mortality responses of *Rhyzopertha dominica* (F.) adults subjected to short exposures on spinosad-treated wheat. Journal of Stored Products Research, 48: 149-152.

GHIMIRE, M.N. ARTHUR, F.H., MYERS, S.M., PHILLIPS, T.W. 2016. Residual efficacy of deltamethrin and β-cyfluthrin against *Trogoderma variabile* and *Trogoderma inclusum* (coleopteran: Dermestidae). Journal of Stored Products Research, **66**:6-11.

MERTZ, E.P., YAO, R.C., 1990. *Saccharopolyspora spinosa* sp nov isolated from soil collected in a sugar rum still. International Journal of Systemic Bacteriology. **40**:34-39.

PHILLIPS, T.W., THRONE, J.E., 2010. Biorational approaches to managing stored-product insects. Annual Review of Entomology. **55**:375-397.

SALGADO, V.L., SPARKS, T.C., 2005. The spinosyns: Chemistry, biochemistry, mode of action, and resistance. In: Gilbert. L.I., Latrou, K., Gill, S.S. (Eds.), Comprehensive Molecular Insect Science, Vol. 6, Elsevier, Oxford, 137-173.

SUBRAMANYAM, B., HARTZER, M., BOINA, D.R., 2013. Performance of pre-commercial release formulations of spinosad against five stored-product insect species on four stored commodities. Journal of Pest Science 85: 331-339.

WIJAYARATNE, L. K. W. AND R. H. S. RAJAPAKSE 2018. Effects of spinosad on the heat tolerance and cold tolerance of *Sitophilus oryzae* L. (Coleoptera: Curculionidae) and *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae). Journal of Stored Products Research 77: 84-88.

WIJAYARATNE, L. K. W., ARTHUR, F.H., WHYARD, S., 2018. Methoprene and control of stored-product insects. Journal of Stored Products Research, **76**:161-169

Effectiveness of spinosad and spinetoram against five stored-product beetle pests under high relative humidity conditions

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The objective of this study was to evaluate spinosad and spinetoram effectiveness against *S. granarius, S. oryzae*, *T. confusum*, *T. castaneum* and *R. dominica* in wheat grain under high relative humidity (75%). The insecticides were applied at the rates of 0.5, 1 and 2 mg a.i./kg. Mortality was assessed after 2, 7, 14 and 21 days, and progeny reduction and grain damage caused by the insects were also assessed.

All rates of both insecticides caused 98-100% mortality of *R. dominica* after 7 days, and 100% mortality after 14 and 21 days of exposure. Both insecticides achieved high mortality (97-100%) after 21 days of contact of *S. granarius* with 1 and 2 mg/kg, and *S. oryzae* with 2 mg/kg rate. The highest mortality of *T. confusum* and *T. castaneum* was recorded after 21 days of contact with 2 mg/kg of both insecticides, 54-55% and 25-31%, respectively. All rates of both insecticides caused high progeny reduction of >99% of *R. dominica*, >90% of *T. confusum* and 94% of *T. castaneum* (only with 2 mg/kg). The highest *S. granarius* progeny reduction (>90%) was found in wheat treated with 2 mg/kg spinosad and 1-2 mg/kg spinetoram, while the greatest progeny reduction of *S. oryzae* was observed in wheat treated with 2 mg/kg spinetoram. Wheat grain damage caused by *R. dominica* was very low, i.e. up to 0.2% in wheat treated with all rates of spinosad and spinetoram, while *S. granarius* and *S. oryzae* caused up to 5% damage only in wheat treated with 2 mg/kg of spinetoram.

Keywords: stored-product beetle pests; high relative humidity; spinosad; spinetoram; effectiveness

1. Introduction

Traditional grain protecting organophosphates (OP) and pyrethroids (PY) still constitute the most important segment in the concept of IPM programs and the first option for control of stored insects in storages in which fumigation is not possible (Arthur 2012; Arthur and Subramanyam, 2012). Further use of these insecticides is limited by resistance that populations of stored-product insects have developed to traditional grain protectants and especially by an increasing consumer demand for products that are free of insects and insecticide residues and their negative impact on the environment (Phillips and Throne, 2010; Boyer et al., 2012). Dealing with the challenge and improvement of anti-resistance tactics and control programs for stored-product insects becomes possible with new insecticides that have different mechanisms of activity and good toxicological and ecotoxicological profiles (Phillips and Throne, 2010; Hertlein et al., 2011). Over the past 20 years, only spinosad and diatomaceous earths have proved to be good alternative to OPs and PYs and have been registered as grain protectants in many countries (Arthur and Subramanyam, 2012; Kljajić et al., 2014). Spinosad is a broad-spectrum insecticide of low mammalian toxicity, a mixture of spinosyn A and spinosyn D, secondary metabolites of the soil actinomycete *Saccaropolyspora spinosa* Mertz and Yao (Hertlein et al., 2011). Spinetoram is a new member of the spinosyn group, and a mixture of two synthetically modified spinosyns (spinosyn J and spinosyn L), which are also metabolites of *S. spinosa*. In the latest research, spinetoram has shown similar or higher effectiveness than spinosad against stored-product insects, and may therefore be expected to play an important role in future control of stored-product insects (Vassilakos et al., 2012; Vassilakos and Athanassiou, 2013; Athanassiou and Kavallieratos, 2014; Vassilakos et al., 2015; Rumbos et al., 2018). Unlike traditional grain protectants, such as OPs that achieve their high efficacy after exposure periods of 2-7 days (Kljajić and Perić 2009; Rumbos et al., 2013) spinosad and spinetoram show slower activity, reaching peak efficacy against most stored-product insects after 14-21 days (Fang et al., 2002; Nayak et al., 2005; Athanassiou et al., 2008; Vayias et al., 2009; Andrić et al., 2011; Vassilakos et al., 2012; Vassilakos and Athanassiou, 2013; Vassilakos et al., 2015; Rumbos et al., 2018). As a result, environmental conditions, such as temperature and relative humidity, may significantly affect the efficacy of spinosad and spinetoram. While higher temperature usually increases efficacy, high humidity mostly leads to efficacy reduction (Athanassiou et al., 2008; Vassilakos and Athanassiou, 2013). Besides reducing insecticide efficacy, high relative humidity most often has a positive effect on progeny production, insect distribution and abundance (Hagstrum et al., 1996). High humidity, especially when it extends over a longer period of time, also leads to greater grain moisture, so that wheat grains become softer and more prone to insect infestation (Gaines et al., 1996).

To our knowledge, the efficacy of spinetoram against the granary weevil *Sitophilus granarius* (L.) and red flour beetle *Tribolium castaneum* (Herbst) under high relative humidity conditions has not been tested so far. The intention of this study was to examine and compare the efficacies of spinosad and spinetoram against *S. granarius*, rice weevil *Sitophilus oryzae* (L.), lesser grain borer *Rhyzopertha dominica (F.)*, confused flour beetle *Tribolium confusum* (Du Val) and *T. castaneum* under high relative humidity (75%), as well as their effects on progeny production/reduction in F_1 generation, and grain damage.

2. Materials and Methods

Test Insects and insecticides used

Laboratory populations of *S. granarius, S. oryzae*, *T. confusum*, *T. castaneum* and *R. dominica*, reared in an insectary, were used in the testing, and procedures described by Harein and Soderstrom (1966), and Davis and Bry (1985) were employed. *S. granarius* and *S. oryzae* were reared in 2.5 L glass jars containing whole-grain soft wheat with moisture content below 12%, while coarse ground wheat was added for *R. dominica* and *T. confusum,* and *T. castaneum* was reared on white wheat flour with 5% yeast. Air temperature in the insectary was 25±1ºC, and relative humidity 60±5 %. Unsexed 2-4 week old adults of all tested species were used in the experiment.

The following commercial products were used in the experiment: Laser 240 SC containing 240 g/L spinosad, and Delegate 250 WP containing 250 g/kg spinetoram (Dow AgroSciences, Austria).

Bioassays

Investigation was conducted in the laboratory under high relative humidity conditions of 75±5% and 25±1ºC temperature (both parameters were measured by a data logger Kestrel 4000, USA). Moisture content in wheat grain, variety 'Simonida', was 12.3±0.1 % and it was measured by a Dickey–John Mini GAC (Dickey–John Co., USA) device before the experiment.

Two standard solutions were prepared for both insecticide and diluted into dose series of 0.5, 1.0 and 2.0 mg a.i./kg, so that each insecticide dose was used for two treatments of 500 g lots. Each 1000 mL glass jar was filled with 500 g of wheat grain and treated with 5 mL of water solution of one of the insecticides, or 5 mL of water for control grain. After hand shaking the treated wheat for 30 s, each jar was placed on a mechanical roller for 15 minutes. For each tested species, six 50 g samples (three per treatment), representing each dose and insecticide, were placed into 200 mL plastic vessels. The next day, 25 adults of each tested species were released into each vessel, which was then covered with cotton cloth and fixed with rubber band. Adult mortality of the tested species was determined 2, 7, 14 and 21 days after the beginning of their exposure to treated wheat grain.

After the last assessment, dead and living adults were removed and the vessels were retrieved to the laboratory (25±1°C and 75±5% r.h.) for additional periods of 5 weeks for *Sitophilus* species, 7 weeks for *R. dominica* and 9 weeks for *Tribolium* species. Progeny emergence/suppression was determined by counting adults (for *Tribolium* species the total number included larvae, pupae and adults).

When the progeny were counted, damage caused by the weevils and *R. dominica* were also assessed on 100 randomly selected kernels per vessel.

Data analysis

Before analysis, percentage mortality was transformed using *arcsine* and progeny counts were transformed by *log(x+1)*. All data were submitted to a one-way ANOVA and the means were separated by Fisher's LSD test at P=0.05. Progeny reduction (IR%) in wheat grain was determined using a formula recommended by Tapondjou et al. (2002).

3. Results

All application rates of spinosad and spinetoram caused low mortality (0-16%) of all tested species after 2 days of exposure, except of *R. dominica*, whose mortality was 18.0-62.7% and 32.7-53.3%, respectively. Seven to 21 days exposure to both insecticides at all application rates caused high mortality of *R. dominica*, 98-100%. The 7 days exposure period to all rates of spinosad and spinetoram caused *S. granarius* mortality that ranged from 18.0-81.3% and from 62.0-86.0%, considerably lower mortality of *S. oryzae* (8.7-72.0% and 24.0-87.3%), and the least (<12%) in *T. confusum* and *T. castaneum*. After 14 and 21 days, spinosad and spinetoram caused high mortality (92-100%) of *S. granarius* and *S. oryzae* in contact with 1.0 and 2.0 mg/kg, and 2.0 mg/kg of both insecticides, respectively, while the highest mortality of *T. confusum* and *T. castaneum* was recorded after 21 days of contact with 2.0 mg/kg of both insecticides, 54-55.3% and 25.3-31.3%, respectively (Table 1).

All application rates of both insecticides caused high progeny reduction: 98.6-100% of *R. dominica*, 90-100% of *T. confusum* and 94.5% of *T. castaneum* (only with 2.0 mg/kg). The highest *S. granarius* progeny reduction (90.2-95.3%) was found in wheat treated with 2 mg/kg spinosad and 1-2 mg/kg spinetoram, while *S. oryzae* progeny reduction was the greatest (94.5%) in wheat treated with 2 mg/kg spinetoram (Table 2).

No grain damage caused by *R. dominica* was detected in wheat treated with any rate of spinosad or spinetoram other than grain treated with the lowest doses of the insecticides, and even that dose caused only a very small damage of up to 0.2%. After *S. granarius* and *S. oryzae* progeny were counted in all treated wheat, grain damage was detected, peaking with 42.2-77.2% in wheat treated with 0.5 mg/kg spinosad, while the lowest was 3.0-5.2%, found in wheat treated with 2 mg/kg spinetoram (Table 2).

4. Discussion

The results in our present study show that the efficacy of spinosad and spinetoram depend on the rate, exposure interval and target species, which is consistent with previous results (Fang et al., 2002; Nayak et al., 2005; Subramanyam et al., 2007; Athanassiou et al., 2008; Vassilakos et al., 2012; Vassilakos and Athanassiou, 2013). For example, both insecticides were highly effective at the rate of 0.5 mg/kg against *R. dominica* after 7 days, at the rate of 1-2 mg/kg against *S. granarius*, and 2 mg/kg rate against *S. oryzae* after 14 days, while the 2 mg/kg rate achieved its highest efficacy against *T. confusum* and *T. castaneum* after 21 days.

In a recent study Athanassiou and Kavallieratos (2014) concluded that spinetoram was equally and in some cases even more effective than spinosad against stored-product beetle species. Based on these results, we can similarly conclude that no significant difference emerged in our experiment between spinosad and spinetoram effectiveness against the most susceptible *R. dominica* and least susceptible *Tribolium* species.

Regarding *Sitophilus* species, however, spinetoram was significantly more effective than spinosad. For example, spinetoram applied at the rates of 0.5 and 1 mg/kg was significantly more effective than spinosad after 7-21 days of exposure of *S. granarius* and *S. oryzae*, as well as considering the average progeny counts and percentage of damaged grain, which was significantly lower for both species in wheat treated with spinetoram.

A number of previous studies (Fang et al., 2002; Nayak et al., 2005; Subramanyam et al., 2007; Athanassiou et al., 2008; Vayias et al., 2009, 2010; Vassilakos et al., 2012, 2015; Athanassiou and Kavallieratos, 2014; Rumbos et al., 2018) agreed that *R. dominica* was the most susceptible of stored-product beetle species, while *Sitophilus* species were significantly less susceptible, and *Tribolium* species the least susceptible to spinosad and spinetoram, which was further confirmed in our present research under high relative humidity conditions. Furthermore, our results clearly show that *S. granarius* is significantly more susceptible than *S. oryzae* to both insecticides, while *T. confusum* is significantly more susceptible than *T. castaneum*. Differences between *Sitophilus* species were greatest when 0.5 mg/kg rate was applied, and after exposure periods of 7 and 14 days, as well as between *Tribolium* species after the application of 2 mg/kg spinosad and spinetoram and exposure for 21 days.

Tab. 1 Mean (% ± SE) mortality of *S. granarius*, *S. oryzae*, *R. dominica*, *T. confusum* and *T. castaneum* adults exposed for 2, 7, 14 and 21 days to wheat treated with spinosad or spinetoram (for each species/exposure separately, means within columns marked by the same letter are not significantly, Fisher's LSD test at *P*>0.05)

Insecticide	Rate mg/kg	Mean (% ± SE) mortality after exposure			
		2 days	**7 days**	**14 days**	**21 days**
S. granarius					
Spinosad	**2.0**	0.7±0.2 a	81.3±0.8 a	99.3±0.2 a	99.3±0.2 a
	1.0	0.0±0.0 a	68.0±1.3 b	98.0±0.3 a	100±0.0 a
	0.5	0.0±0.0 a	18.0±1.0 c	48.7±1.8 c	71.0±1.1 c
Spinetoram	**2.0**	0.0±0.0 a	86.0±0.6 a	100±0.0 a	100±0.0 a
	1.0	0.0±0.0 a	80.0±0.6 a	100±0.0 a	100±0.0 a
	0.5	0.0±0.0 a	62.0±1.2 b	88.0±05 b	94.0±0.7 b
S. oryzae					
Spinosad	**2.0**	4.0±0.4 b	72.0±1.1 b	92.0±0.8 ab	97.3±0.3 a
	1.0	3.3±0.3 bc	42.0±1.1 c	60.0±1.0 c	71.0±0.8 b
	0.5	0.0±0.0 c	8.7±0.8 d	18.7±1.2 d	21.3±12 d
Spinetoram	**2.0**	16.0±0.4 a	87.3±0.7 a	98.0±0.5 a	98.7±03 a
	1.0	3.3±0.3 bc	52.0±0.9 c	87.3±1.4 b	96.0±0.4 a
	0.5	0.0±0.0 c	24.0±1.4 d	36.7±1.2 d	44.7±1.3 c
R. dominica					
Spinosad	**2.0**	62.7±0.4 a	100±0.0 a	100±0.0 a	100±0.0 a
	1.0	42.7±0.6 c	100±0.0 a	100±0.0 a	100±0.0 a
	0.5	18.0±0.8 e	98.0±0.2 b	100±0.0 a	100±0.0 a
Spinetoram	**2.0**	53.3±0.5 b	100±0.0 a	100±0.0 a	100±0.0 a
	1.0	48.7±1.1bc	100±0.0 a	100±0.0 a	100±0.0 a
	0.5	32.7±0.6 d	99.3±0.2 a	100±0.0 a	100±0.0 a
T. confusum					
Spinosad	**2.0**	0.0±0.0 a	12.0±1.0 a	34.7±2.2 a	54.0±2.2 a
	1.0	0.0±0.0 a	1.3±0.2 b	2.7±0.3 b	6.0±0.6 b
	0.5	0.0±0.0 a	0.0±0.0 b	3.3±0.3 b	5.3±0.3 b
Spinetoram	**2.0**	0.0±0.0 a	10.0±0.4 a	32.7±0.9 a	55.3±1.1 a
	1.0	0.0±0.0 a	2.0±0.2 b	6.7±0.3 b	16.0±0.4 b
	0.5	0.0±0.0 a	0.0±0.0 b	6.7±0.4 b	14.7±0.8 b
T. castaneum					
Spinosad	**2.0**	0.0±0.0 a	9.3±0.6 ab	22.0±1.8 a	25.3±2.3 a
	1.0	0.0±0.0 a	4.7±0.4 bc	9.3±0.5 b	11.3±0.7 b
	0.5	0.0±0.0 a	0.0±0.0 c	4.0±0.2 b	7.3±0.5 b
Spinetoram	**2.0**	0.0±0.0 a	12.0±0.6 a	24.0±0.9 a	31.3±1.0 a
	1.0	0.0±0.0 a	2.0±0.2 c	8.0±0.4 b	10.0±0.4 b
	0.5	0.0±0.0 a	0.0±0.0 c	7.3±0.5 b	10.0±0.6 b

Vassilakos et al. (2012) reported a 1.5 times lower efficacy of 0.5 mg/kg rate of spinetoram in wheat against *S. oryzae* than *S. granarius*. Our study showed even greater differences, so that spinetoram rate of 0.5 mg/kg after 7 days and spinosad rate of 0.5 mg/kg after 14 days were 2.6 times less effective against *S. oryzae* than against *S. granarius*. After 21 days of contact with 2 mg/kg rate of spinosad and spinetoram, efficacy was 1.8 and 2.1 times lower against *T. castaneum* than against *T. confusum*.

Data from some earlier studies show that increasing relative humidity mostly leads to lower efficacy of spinosad and spinetoram (Athanassiou et al., 2008; Vassilakos and Athanassiou, 2013). Comparing efficacy data for spinosad under high humidity of 75% in the present study and our earlier findings (Andrić et al., 2011) in experiments conducted under 60% r.h., similar conclusions were drawn. For example, spinosad applied in our present study at 0.5, 1 and 2 mg/kg rates resulted in *S. oryzae* mortality of 18, 60 and 92% after 14 days, while the respective data from earlier experiments were 59, 77 and 100%. Similarly, progeny reduction of *S. oryzae* in wheat treated with 0.5, 1 and 2 mg/kg spinosad was 2.5, 34.1 and 82.2% at 75% r.h., which is significantly less than progeny reduction at 60% r.h, which was 42.2, 80,8 and 91%.

Tab. 2 Progeny emergence (adults/vessel ± SE), progeny reduction (%) and kernel damage (mean % ± SE) of *S. granarius*, *S. oryzae*, *R. dominica*, *T. confusum* and *T. castaneum* in wheat treated with spinosad or spinetoram (for each species separately, means within columns followed by the same letter are not significantly different, Fisher's LSD test at *P*>0.05)

Insecticide	Rate mg/kg	Progeny emergence (adults/vial ± SE)	Progeny reduction (%)	Kernel damage (mean % ± SE)
S. granarius				
Spinosad	**2.0**	54.3±18.0 e	90.2	13.0±2.8 de
	1.0	87.3±6.7 d	84.4	18.2±2.4 d
	0.5	377.2±13.4 b	33.1	42.2±2.7 b
Spinetoram	**2.0**	25.7±2.0 f	95.3	3.0±0.7 f
	1.0	43.3±7.2 e	92.2	8.5±1.2 ef
	0.5	169.8±19.0 c	69.8	26.8±2.7 c
	0	564.2±33.0 a	-	58.7±3.8 a
S. oryzae				
Spinosad	**2.0**	134.5±35.2 d	82.2	14.7±3.3 c
	1.0	499.2±22.1 b	34.1	51.0±3.1 b
	0.5	738.5±16.9 ab	2.5	77.2±3.5 a
Spinetoram	**2.0**	41.8±.4.2 e	94.5	5.2±0.7 d
	1.0	196.2±14.0 c	74.1	21.8±2.3 c
	0.5	511.0±41.6 b	32.5	51.2±4.0 b
	0	757.5±34.0 a	-	76.0±1.3 a
R. dominica				
Spinosad	**2.0**	0.0±0.0 b	100	0.0±0.0 b
	1.0	0.0±0.0 b	100	0.0±0.0 b
	0.5	2.2±0.4 b	98.6	0.2±0.2 b
Spinetoram	**2.0**	0.0±0.0 b	100	0.0±0.0 b
	1.0	0.0±0.0 b	100	0.0±0.0 b
	0.5	0.5±0.3 b	99.3	0.2±0.2 b
	0	238.5±42.5 a	-	29.0±0.03 a
T. confusum				
Spinosad	**2.0**	0.0±0.0 b	100	/
	1.0	0.0±0.0 b	100	/
	0.5	0.0±00 b	100	/
Spinetoram	**2.0**	0.0±00 b	100	/
	1.0	0.0±00 b	100	/
	0.5	0.2±0.2 b	90.0	/
	0	1.7±0.9 a	-	/
T. castaneum				
Spinosad	**2.0**	1.2±0.3 e	94.5	/
	1.0	3.5±0.6 d	84.7	/
	0.5	11.3±2.4 b	51.8	/
Spinetoram	**2.0**	1.2±0.6 e	94.5	/
	1.0	5.3±1.2 cd	77.0	/
	0.5	9.0±17 bc	61.6	/
	0	23.7±3.2 a	-	/

As data from tests of the effectiveness of insecticides as grain protectants may vary, it is very important to determine their effects on progeny production of storage insects, as well as on grain damage (Subramanyam and Roesli 2000; Subramanyam et al., 2007). In the present study, all spinosad and spinetoram doses caused high progeny reduction (98.6-100%) only for *R. dominica*, accompanied by almost no grain damage at all (≤0.2%), as well as high progeny reduction (90-100%) for *T. confusum*, which is consistent with earlier reports (Fang et al., 2002; Vayias et al., 2009; Subramanyam et al., 2007; Athanassiou and Kavallieratos, 2014). Regarding both *Sitophilus* species, however, only the 2 mg/kg rate of spinosad and spinetoram caused high progeny reduction of 82.2-90.2% and 94.5-95.3%, respectively, while the percentage of grain damage ranged 3-14.7% and 3-5.2%, respectively. These results are inconsistent with several earlier studies (Vayias et al., 2009; Subramanyam et al., 2007; Athanassiou and Kavallieratos, 2014) in which 1 mg/kg rate of spinosad or spinetoram applied to various types of grain resulted in high or maximum reduction of progeny of maize weevil *Sitophilus zeamais* (Motsch.), *S. granarius* and *S. oryzae*, and no maize grain damage was caused by *S. zeamais* and *S. oryzae*. Besides the lower effiacy that was observed in this study and population origin, the observed differences may also be attributed to the high relative humidity that made wheat grain softer in our experiment (Gaines et al., 1996). It enabled progeny production of *Sitophilus* species, and consequent grain damage, as well as progeny production of *T. castaneum* even after wheat grain treatment with 2 mg/kg spinosad or spinetoram. Supporting these findings are data on grain damage caused by *S. granrius* and *S. oryzae* in control of 58.7 and 76.0%, respectively.

Based on all results in this study, we concluded that the minimum effective dose of spinosad and spinetoram for *R. dominica* and *S. granarius* control in wheat grain under high humidity conditions (75% r.h.) is 0.5 and 2.0 mg/kg, respectively, and only 2 mg/kg of spinetoram for *S. oryzae* control, while successful control of *T. confusum* requires 0.5 mg/kg of spinosad or spinetoram, and *T. castaneum* 2.0 mg/kg of either spinosad or spinetoram.

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References

ANDRIĆ, G., KLJAJIĆ, P. UND M. PRAŽIĆ GOLIĆ, 2011: Effects of Spinosad and Abamectin on different Populations of Rice Weevil *Sitophilusoryzae* (L.) in Treated Wheat Grain. Pesticides and Phytomedicine **26**, 377-384.

ARTHUR, F.H., 2012: Aerosols and contact insecticides as alternatives to methyl bromide in flour mills, food production facilities, and food warehouses. Journal of Pest Science **85**, 323-329.

ARTHUR, F.H. UND BH. SUBRAMANYAM, 2012: Chemical control in stored products. In Hagstrum, D.W., Philips, T.W. & Cuperus, G. (Eds.), Stored product protection (pp 95-100). Kansas State University Agricultural Experiment Station and Cooperative Extension Service. Retrieved from https://www.bookstore.ksre.ksu.edu/pubs/s156.pdf

ATHANASSIOU, C.G UND N.G. KAVALLIERATOS, 2014: Evaluation of spinetoram and spinosad for control of *Prostephanus truncatus*, *Rhyzopertha dominica*, *Sitophilus oryzae*, and *Tribolium confusum* on stored grains under laboratory tests. Journal of Pest Science **87**, 469-483.

ATHANASSIOU, C.G., KAVALLIERATOS, N.G., YIATILLIS, E., VAYIAS, B.J., MAVROTAS, S.C. UND Ž. TOMANOVIĆ, 2008: Influence of temperature and humidity on the efficacy of spinosad against four stored-grain beetle species. Journal of Insect Science **8**, 1-9.

BOYER, S., ZHANG, H. UND G. LEMPERIERE, 2012: A review of control methods and resistance mechanisms in stored-product insects. Bulletin of Entomological Research **102**, 213-229.

DAVIS, R. UND R.E. BRY, 1985: *Sitophilus granarius, Sitophilus oryzae* and *Sitophilus zeamais; Tribolium confusum* and *Tribolium castaneum*. In: Handbook of Insect Rearing (Eds. Singh, P., Moore, R.F.), Elsevier, Amsterdam-Oxford-NewYork-Tokyo, 287-293.

FANG, L., SUBRAMANYAM, BH. UND F.H. ARTHUR, 2002: Effectiveness of spinosad on four classes of wheat against five stored-product insects. Journal of Economic Entomology, **95**, 640-650.

GAINES, C.S., FINNEY, P.F., FLEEGE, L.M. AND L.C. ANDREWS, 1996: Predicting a hardness measurement using the single-kernel characterization system. Cereal Chemistry, **73**, 278-283.

HAGSTRUM, D.W., FLINN, P.W. UND D.W. HOWARD, 1996: Ecology. In: Integrated Management of Insects in Stored Products. (Eds. Subramanyam, B., Hagstrum, D.W.), Marcel Dekker, Inc., New York-Basel-Hong Kong, 71-134.

HAREIN, C.R. UND E.L. SODERSTROM, 1966: Coleoptera infesting stored products. In: Insect Colonization and Mass Production (Ed. Smith, C.N.), Academic Press, New York and London, 241-257.

HERTLEIN, M.B., THOMPSON, G.D., SUBRAMANYAM, BH. UND C.G. ATHANASSIOU, 2011: Spinosad: A new natural product for stored grain protection. Journal of Stored Products Research, **47**, 131-146.

KLJAJIĆ, P., KAVALLIERATOS, N.G., ATHANASSIOU, C.G. UND G. ANDRIĆ, (2014): Is combining different grain protectans a solution to problems with resistant populations of stored-product insects? Proceedings of the 10th International Working Conference on Stored Product Protection, 24-28 November 2014 Chiang Mai, Thailand, pp. 781- 793.

KLJAJIĆ, P. UND I. PERIĆ, 2009: Residual effects of deltamethrin and malathion on different populations of *Sitophilus granarius* (L.) on treated wheat grains. Journal of Stored Products Research, **45**, 45-48.

NAYAK, M.K., DAGLISH, G.J. UND V.S. BYRNE, 2005: Effectiveness of spinosad as a grain protectant against resistant beetle and psocid pests of stored grain in Australia. Journal of Stored Products Research, **41**, 455-467.

PHILLIPS, T.W. UND J.E. THRONE, 2010: Biorational approaches to managing stored-product insects. Annual Review of Entomology **55**, 375-397.

RUMBOS, C.I., DUTTON, A.C. UND C.G. ATHANASSIOU, 2013: Comparison of two pirimiphos-methyl formulations against major stored-product insect species. Journal of Stored Product Research **55,** 105-106.

RUMBOS, C.I., DUTTON, A.C. UND C.G. ATHANASSIOU, 2018: Insecticidal effect of spinetoram and thiamethoxam applied alone or in combination for the control of major stored-product beetle species. Journal of Stored Products Research **75**, 56-63.

SUBRAMANYAM, BH., UND D.W. HAGSTRUM, 1996: Resistance measurement and management. In: Integrated Management of Insects in Stored Products. (Eds. Subramanyam, B., Hagstrum, D.W.), Marcel Dekker, Inc., New York-Basel-Hong Kong, 331-397.

SUBRAMANYAM, BH. UND R. ROESLI, 2000: Inert dusts In: Alternatives to Pesticides in Stored-Product IPM (Eds. Subramanyam, B., Hagstrum, D.W.), Kluwer Academic Publishers, Boston/Dordrecht/London, 321-380.

SUBRAMANYAM, BH., TOEWS, M.D., ILELEJI, K.E., MAIER, D.E., THOMPSON, G.D. UND T.J. PITTS, 2007: Evaluation of spinosad as a grain protectant on three Kansas farms. Crop Prot. **26**, 1021-1030.

TAPONDJOU, L.A., ADLER, C., BOUDA, H. UND D.A. FONTEM, 2002: Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles. Journal of Stored Products Research **38**, 395-402.

VASSILAKOS, T.N. UND C.G. ATHANASSIOU, 2013: Effect of temperature and relative humidity on the efficacy of spinetoram for the control of three stored product beetle. Journal of Stored Products Research **55**, 73-77.

VASSILAKOS, T.N. ATHANASSIOU, C.G. SAGLAM, O., CHLORIDIS, A.S. UND J.E. DRIPPS, 2012: Insecticidal effect of spinetoram against six major stored grain insect species. Journal of Stored Products Research **51**, 69-73

VASSILAKOS, T.N. ATHANASSIOU, C.G. UND N.G. TSIROPOULOS, 2015: Influence of grain type on efficacy of spinetoram for control of *Rhyzopertha dominica*, *Sitophilus granarius* and *Sitophilus oryzae*. Journal of Stored Products Research **64**, 1-7.

VAYIAS, B.J., ATHANASSIOU, C.G., MILONAS, D.N. UND C. MAVROTAS, 2009: Activity of spinosad against three stored-product beetle species on four grain commodities. Crop Protection, **28**, 561-566.

VAYIAS, B.J., ATHANASSIOU, C.G., MILONAS, D.N. UND C. MAVROTAS, 2010: Persistence and efficacy of spinosad on wheat, maize, and barley grains against four major stored product pests. Crop Protection, **29**, 496-505.

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Spinosad-induced stress on the maize weevil *Sitophilus zeamais*

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Abstract

Although seldom considered, sublethal insecticide exposure may lead to harmful, neutral, or even beneficial responses that may affect (or not) the behavior and fitness of the exposed insects. Intriguingly, little is known about such effects on stored product insect pests and even less is available regarding the bioinsecticide, spinosad. Thus, we assessed the sublethal effects of spinosad on walking, feeding, drinking and mating behaviors of maize weevils (*Sitophilus zeamais*), also assessing their survival, reproductive output, and grain loss compared with maize weevils exposed to the pyrethroid deltamethrin (as positive control), and water only (negative control). Both spinosad and deltamethrin were able to effectively control the insects, although the latter caused a faster mortality than the former. Behavioral pattern changes were caused by both insecticides, especially deltamethrin, triggering irritability (i.e., avoidance after contact). Different feeding and drinking responses were also detected with significant avoidance to deltamethrin, but not to spinosad. Maize weevil couples sublethally exposed to deltamethrin and spinosad exhibited altered reproductive behavior, a likely consequence of their altered activity, but deltamethrin caused greater behavioral changes. Curiously, higher progeny emergence and grain loss were observed in deltamethrin-exposed insects, suggesting that this pyrethroid insecticide elicits hormesis in maize weevils that may compromise control efficacy by this compound. In contrast, such effect was not detected with spinosad, which did not elicit avoidance allowing the intended weevil exposure and control.

Keywords: biopesticide, hormesis, insecticide avoidance, sublethal exposure, progeny production

Introduction

Insecticides are a familiar class of pest control agents, understandable due to their broad use since the 1940's across many sectors, including stored product protection. Although insecticides are technically defined as "*any substance or mixture of substances intended for preventing, destroying, repelling, or mitigating any insect pest*" (e.g., US Federal Insecticide, Fungicide, and Rodenticide Act), these compounds are popularly defined as substances that kills insects. The blame probably lays with the old Romans and the Latin origin of the suffix *cide* (from *cīda*; = a killer of), which is rather popular and frequent in several nouns of different languages. Regardless, the emphasis of the popular definition of insecticide is on the *killing* of insects, not managing or controlling them, as