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Effects of Spinosad and Abamectin on different Populations of Rice Weevil *Sitophilus oryzae* (L.) in Treated Wheat Grain

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SUMMARY

In laboratory conditions (25±1°C and 60±5% r.h.) effects of natural insecticides spinosad and abamectin on five *S. oryzae* populations (laboratory, Šid, Gornji Milanovac, Žabari and Novi Pazar) were investigated. Both insecticides for all tested populations were applied to untreated wheat grain at following rates 0.25, 0.5, 1.0 and 2.0 mg AI/kg, subsequently 25 adults were added in each plastic vessels (V=200 cm^3) containing 50 g of treated wheat, in four replicates, for each population tested. Mortality of weevils was determined after 2-, 7- and 14-days, and the effect on progeny production was determined 8-weeks from parental exposure.

Efficacy of spinosad and abamectin after 2-days of weevil exposure for all tested populations and all application rates was <15%. After 7-days of exposure, the efficacy was ≥95% for weevils from Žabari, in wheat treated with 2 mg/kg of spinosad and abamectin, and for weevils from Gornji Milanovac, only in wheat treated with 2 mg/kg of spinosad. After 14-days of exposure the efficacy ≥95% was found for laboratory weevils and weevils from Žabari and Gornji Milanovac, in wheat treated with 1 and 2 mg/kg of spinosad, and for *S. oryzae* from Novi Pazar and Šid, in wheat treated with 2 mg/kg of spinosad. At the same time for all tested populations abamectin at rates of 0.5, 1.0 and 2.0 mg/kg was 94-100% efficient. No insecticide achieved total (100%) progeny reduction in tested populations of *S. oryzae,* while high progeny reduction (≥95%) was found only in weevils which were in contact with wheat treated with 1 and 2 mg/kg of abamectin. The results showed that for highly efficient control of different populations of *S. oryzae* in wheat grain, ≥2 mg/kg of abamectin, and, particularly, spinosad should be applied.

Keywords: *S. oryzae;* Wheat grain; Spinosad; Abamectin; Effects

INTRODUCTION

Rice weevil *Sitophilus oryzae* (L.), is very harmful, primary pest of stored wheat worldwide (Rees, 2004). In Serbia, in the last years, this species has become dominant compared to other species of *Sitophilus* genus and it is present in almost all storage facilities (Almaši, 2008).

Chemical control, use of contact insecticides and fumigants, is still the most effective segment of integral pest management in stored products (Kljajić, 2008). Contact insecticides are used for treatment of storage space and/or direct treatment of products ensuring a long-term protection from all storage insects. Although the selection of insecticides for this purpose has changed over time, organophosphorus and pyrethroid compounds are still predominantly used in control of *S. oryzae* and other storage pests worldwide (White and Leesch, 1996; Tomlin 2009). In Serbia, six products based on four active ingredients are registered for this purpose: dichlorvos, malathion, pirimiphos-methyl and deltamethrin (Kljajić, 2008; Janjić and Elezović, 2010). However, it is known that altered susceptibility of storage insects to contact insecticides is the most important limiting factor for their use, because, apart from the lack of pest control effects, application rates are increased which results in presence of harmful residues in food and adversely affects food quality, human health and the environment (Subramanyam and Hagstrum, 1996; Kljajić and Perić, 2005).

To overcome these problems, in recent years new insecticides for storage insects control are being investigated, and special attention is payed to insecticides with natural origin, such as spinosad and abamectin. Spinosad is a mixture of spinosyn A and spinosyn D, secondary metabolites of soil actinomycete *Saccharopolyspora spinosa* (Thompson et al., 2000; Copping and Duke, 2007; Krämer and Schirmer, 2007). It is registered for control of a number of insects from orders Lepidoptera, Diptera, Thysanoptera and Coleoptera, in over 100 crops. Since 2005 in USA and some African countries it has been registered for use in storages at rate of 1 mg AI/kg (Subramanyam, 2006), and due to favorable toxicological and ecotoxicological properties it was also aproved for use in organic food production (Hertlein et al., 2011). Among storage insects, *Rhyzopertha dominica* (F.) is the most susceptible to spinosad, even populations which were found to be resistant to organophosphates and pyrethroids (Nayak et al., 2005), and it can be applied in combination with other insecticides of natural origin such as, for instance, diatomaceus earth, in which cases it provides high efficacy at lower rates (Chintzoglou et al., 2008).

Abamectin is a mixture of avermectin B_{1a} and B_{1b}, fermentation products of actinomycete *Streptomyces avermitilis* and is used in protection of many cultivated plants from pest insects and mites (Copping and Duke, 2007; Krämer and Schirmer, 2007), provided that, so far, no product has been registered for use in storage facilities in the world. However, previous studies had shown that abamectin expresses high efficacy in control of *S. oryzae*, *R. dominica*, *Tribolium confusum* Jacquelin du Val and different populations of *Tribolium castaneum* (Herbst) (Kavallieratos et al., 2009; Andrić et al., 2010; Pražić Golić et al., 2010).

The aim of the paper was to determine, under laboratory conditions, the efficacy of spinosad and abamectin against *S. oryzae*, as well as their effects on reproduction, after different exposure periods in treated wheat grain of one laboratory and four populations from storages in Serbia.

MATERIAL AND METHODS

Tested populations and insectides used

Laboratory population of *S. oryzae* and populations collected from the following localities: Šid (silo, in the year 2010), Gornji Milanovac (warehouse, 2010), Žabari (silo, 2009) and Novi Pazar (silo, 2002) were used in the study. All populations were reared in insectarium, according to methods described by Harein and Soderstrom (1966) and Davis and Bry (1985), in 2.5 dm^3 glass jars, on the soft whole grain wheat of 12% humidity, at temperature of 25±1°C and relative air humidity 60±5%. Adult insects aged two to four weeks were used in the experiment, at unknown sex ratio.

The following commercial products were used in the experiment: Spinosad 240 SC (NAF-315) containing 22.8% of spinosad (Dow AgroSciences, USA) and Abastate, EC formulation containing 18g/L of abamectin (Galenika-Fitofarmacija, Serbia).

Bioassays

Investigations were conducted under laboratory conditions (25±1°C and 60±5% r.h.) according to modified method described by Collins (1990) and methods for insecticide efficacy evaluation in storage pests control (OEPP/EPPO, 2004a, 2004b). Untreated soft wheat, variety Apač, with 11.5±0.3% of water, determined by the device Dickey–John Moisture Meter (Dickey–John Mini GAC Dickey–John Co.,USA) was used in the experiment.

For all tested populations, insecticides were applied at the following rates: 0.25, 0.5, 1.0 and 2.0 mg AI/kg, and for each rate 1 kg of wheat was treated (10 mL of insecticide water solution per 1 kg of untreated wheat grains), while control wheat grains were treated with water. After 30 seconds of manual shaking of treated wheat, mixing on rotary stirrer for 10 minutes was performed, and subsequently in 200 cm^3 plastic vessels for each tested population 50 g of treated wheat in four replicates was added. The following day, 25 adults were put in each vessel.

Mortality of individuals from tested populations was determined after 2-, 7- and 14-days from the beginning of their exposure to treated wheat grains, and effect of insecticides to progeny production was determined 8-weeks after parental contact with treated wheat, and untreated wheat in the control.

Data analysis

The acquired mortality data were adjusted for mortality in the control by Abbott formula (1925), and given as percentages. Before analysis, percentage mortality was transformed using *arcsine*, and progeny number was transformed using *log(x+1)*, and then were statistically compared using one-way ANOVA and the significance of mean differences was determined by Fisher's LSD test at $P<0.05$ (Sokal and Rohlf, 1995). However, untransformed means and standard errors are shown in the tables. The reduction in progeny size against the control is shown as a percentage (PR %) according to formula used in similar studies by Tapondjou et al. (2002).

RESULTS

Efficacy of spinosad and abamectin after 2-day exposure for all tested populations was very low (Tables 1-5), and nominally the highest for *S. oryzae* from Žabari (14%) in wheat treated with 2 mg/kg of spinosad. After 7-days of exposure, efficacy >90% was recorded for wheat treated with 2 mg/kg of spinosad, for laboratory weevils (92%), and weevils from Šid (92%), Gornji Milanovac (95%) and Žabari (97%), as well as for weevils from Žabari (99%) in wheat treated with 2 mg/kg of abamectin. In this period, counting all tested populations and both insecticides, spinosad at the lowest rate (0.25 mg AI/kg) achieved the lowest efficacy, 2-17%, while abamectin applied at the same rate achieved significantly higher efficacy, 31-53%.

After 14-days of exposure, in all tested variants, significantly higher efficacy was recorded compared to exposure period of 7-days, therefore, taking into account all tested populations and both insecticides, the most significant increase was recorded for 0.25 mg/kg of abamectin, from 31-53% to 83-94%. After 14-days of exposure efficacy ≥95% was recorded for *S. oryzae* laboratory population and populations from Gornji Milanovac and Žabari, in wheat treated with 1 and 2 mg/kg of spinosad, and for *S. oryzae* from Šid and Novi Pazar, in wheat treated with 2 mg AI/kg. Simultaneously, abamectin at rates 0.5, 1.0 and 2.0 mg/kg was 94-100% efficient against all tested populations.

In all tested variants, the occurrence of progeny of *S. oryzae* was observed (Tables 1-5). All tested rates of abamectin in all tested populations achieved statistically higher progeny reduction than spinosad. In wheat treated with spinosad, progeny reduction >90% was found in laboratory population (91.6%), in weevils from Žabari

Table 1. Effects of spinosad and abamectin on *S. oryzae* from laboratory population

Insecticide	Rate (mg/kg)	Mortality (%±SE) after exposure			Progeny reduction	
		2 days	7 days	14 days	No±SE	PR (%)
Spinosad	2.0	5.0±0.6 a*	92.0±0.8 f	100 f	70.0±13.8 e	91.6
	1.0	0.0±0.0 a	79.0±0.7 e	96.0±0.4 e	160.3±22.8 c	80.8
	0.5	0.0±0.0 a	48.0±2.0 bc	66.0±1.4 c	485.5±35.4 b	42.2
	0.25	0.0±0.0 a	11.0±1.3 a	18.0±1.5 b	602.0±45.5 b	28.4
Abamectin	2.0	3.0±0.2 a	70.0±0.8 de	100 f	12.2±1.5 g	98.4
	1.0	0.0±0.0 a	62.0±1.4 cd	100 f	22.5±2.8 f	97.2
	0.5	0.0±0.0 a	41.0 ±1.5 b	100 f	66.2±2.0 e	92.0
	0.25	0.0±0.0 a	31.0±0.8 b	83.0±0.7 d	96.7±9.3 d	88.4
	0	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	840.7±18.7 a	-

* Means within columns followed by the same letter are not significantly different ($p<0.05$)

Table 2. Effects of spinosad and abamectin on *S. oryzae* population from Šid

Insecticide	Rate (mg/kg)	Mortality (%±SE) after exposure			Progeny reduction	
		2 days	7 days	14 days	No±SE	PR (%)
Spinosad	2.0	4.0±0.6 b*	92.0±1.1 d	100 f	50.2±12.6 c	89.3
	1.0	0.0±0.0 a	63.0±2.3 c	77.0±2.9 c	219.0±53.5 b	54.9
	0.5	0.0±0.0 a	42.0±1.5 b	59.0±5.2 b	408.2±82.4 a	16.3
	0.25	0.0±0.0 a	2.0±0.3 a	2.0±0.6 a	494.5±50.9 a	-1.3
Abamectin	2.0	5.0±0.5 b	88.0±1.2 d	100 f	5.0±1.4 f	98.6
	1.0	0.0±0.0 a	52.0±1.8 bc	100 f	9.2±1.8 e	97.7
	0.5	0.0±0.0 a	63.0±1.1 c	97.0±0.5 e	17.2±1.8 d	96.1
	0.25	0.0±0.0 a	33.0±1.6 b	89.0±0.7 d	41.7±5.8 c	91.1
	0	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	488.2±29.3 a	-

* Means within columns followed by the same letter are not significantly different ($p<0.05$)

Table 3. Effects of spinosad and abamectin on *S. oryzae* population from Gornji Milanovac

Insecticide	Rate (mg/kg)	Mortality (%±SE) after exposure			Progeny reduction	
		2 days	7 days	14 days	No±SE	PR (%)
Spinosad	2.0	2.0±0.3 ab*	95.0±0.5 d	100 f	44.2±7.3 d	92.6
	1.0	0.0±0.0 a	69.0±1.5 c	95.0±0.5 de	179.2±23.5 c	74.3
	0.5	0.0±0.0 a	47.0±1.7 b	69.0±1.1 c	382.2±41.5 b	46.7
	0.25	0.0±0.0 a	10.0±0.9 a	26.0±0.6 b	603.5±34.9 a	16.6
Abamectin	2.0	3.0±0.5 b	74.0±2.2 c	100 f	6.0±0.9 f	97.8
	1.0	1.0±0.2 ab	50.0±0.6 b	99.0±0.2 f	20.7±2.9 e	95.8
	0.5	0.0±0.0 a	48.0±1.2 b	98.0±0.3 ef	49.0±6.5 d	92
	0.25	0.0±0.0 a	32.0±0.7 b	92.0±0.6 d	55.0±7.8 d	91.2
	0	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	725.5±34.0 a	-

* Means within columns followed by the same letter are not significantly different ($p<0.05$)

Table 4. Effects of spinosad and abamectin on *S. oryzae* population from Žabari

Insecticide	Rate (mg/kg)	Mortality (%±SE) after exposure			Progeny reduction	
		2 days	7 days	14 days	No±SE	PR (%)
Spinosad	2.0	14.0±07 b*	97.0±0.5 e	100 e	26.5±2.5 c	92.5
	1.0	0.0±0.0 a	89.0±1.1 d	99.0±0.2 e	84.2±14.1 b	77.8
	0.5	0.0±0.0 a	43.0±1.6 b	64.0±1.1 c	293.7±24.9 a	24.1
	0.25	0.0±0.0 a	17.0±0.7 a	27.0±0.6 b	381.7±40.0 a	1.6
Abamectin	2.0	5.0±0.5 a	99.0±0.2 e	100 e	6.2±0.8 e	97.7
	1.0	0.0±0.0 a	88.0±0.4 d	100 e	15.0±2.6 d	95.5
	0.5	0.0±0.0 a	75.0±0.5 c	98.0±0.5 e	21.7±6.3 cd	93.8
	0.25	0.0±0.0 a	53.0±1.3 b	94.0±0.8 d	50.0±5.0 b	86.5
	0	0.0±0.0 a	0.0±0.0 a	2.0±0.3 a	388.0±79.8 a	-

* Means within columns followed by the same letter are not significantly different ($p<0.05$)

Table 5. Effects of spinosad and abamectin on *S. oryzae* population from Novi Pazar

Insecticide	Rate (mg/kg)	Mortality (%±SE) after exposure			Progeny reduction	
		2 days	7 days	14 days	No±SE	PR (%)
Spinosad	2.0	7.0±0.2 b*	81.0±0.8 e	100 d	100.2±17.7 d	85.2
	1.0	0.0±0.0 a	58.0±1.6 cd	90.0±1.9 c	296.0±73.4 c	59.4
	0.5	0.0±0.0 a	33.0±0.8 b	39.0±1.1 b	463.7±36.3 b	37.2
	0.25	0.0±0.0 a	13.0±0.8 a	18.0±1.4 a	643.7±21.5 ab	13.5
Abamectin	2.0	5.0±0.5 b	64.0±1.2 d	100 d	18.5±2.3 f	96.0
	1.0	0.0±0.0 a	57.0±0.6 cd	100 d	34.5±6.0 e	93.9
	0.5	0.0±0.0 a	42.0±2.6 bc	94.0±0.6 c	71.5±4.3 d	89.0
	0.25	0.0±0.0 a	31.0±1.5 b	91.0±0.7 c	82.5±7.0 d	87.6
	0	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	746.0±16.7 a	-

* Means within columns followed by the same letter are not significantly different ($p<0.05$)

(92.5%) and Gornji Milanovac (92.6%), but only after the highest rate of 2 mg/kg was applied, while all applied rates of abamectin achieved the same level of progeny reduction in the weevils, in populations from Šid (91-99%) and Gornji Milanovac (91-98%). Also, abamectin achieved progeny reduction >90% in wheat treated with 0.5, 1.0 and 2.0 mg/kg, in laboratory weevils (92-98%) and populations from Žabari (94-98%), i.e. with 1 and 2 mg/kg, in population from Novi Pazar (94 and 96%).

DISCUSSION

Results presented in the paper have confirmed the findings of previous studies referring to high efficacy of spinosad and abamectin against storage insects, with an emphasis that exposure duration significantly affects achieved efficacy (Fang et al., 2002; Nayak et al., 2005; Kavallieratos et al., 2009; Andrić et al., 2010; Pražić Golić et al., 2010). According to the results obtained in these researches, for high efficacy of spinosad and abamectin against *S. oryzae* adults exposure period of 14-days is necessary. This can be explained by significantly slower action of natural insecticides compared to pyrethroids and organophosphates (Hertlein et al., 2011). For example, Kljajić and Perić (2009) for different populations of *Sitophilus granarius* (L.) recorded high efficacy (99-100%) of organophosphate malathion applied at rate of 10 mg/kg, already after 2-days of exposure in treated wheat with 2-, 7- and 14-days old deposits, but also high efficacy (98-100%) of pyrethroid deltamethrin applied at rate of 0.5 mg/kg, after 7-days of exposure on 90-days old deposits.

Fang et al. (2002) after 7- and 14-days of *S.oryzae* exposure in soft wheat treated with spinosad (1 mg/kg) recorded efficacy of 69% and 93%, respectively, while Kavallieratos et al. (2009) for the same rate of abamectin after 7- and 14-days of *S. oryzae* exposure recorded efficacy of 40% and 99.7%, respectively at temperature of 20°C, i.e. 100% at 30°C. Similar results were obtained in our researches, whereas, taking into account all populations, spinosad efficacy (1 mg/kg) after 7- and 14-days of exposure was in the range 58-89% and 77-99%, respectively, and the same rate of abamectin in the range 50-88% and 99-100%, respectively.

Little data is available on susceptibility of field populations of storage insects to spinosad and abamectin. Fangeng et al. (2004) found 1.7-2.5.-fold lower susceptibility to spinosad in two field populations of *Plodia interpunctella* (Hübner) and *Cryptolestes ferrugineus* (Stephens) and two-fold higher susceptibility in one population of *R. dominica*. Nayak et al. (2005) recorded 2.4-fold lower efficacy of the spinosad label rate (1 mg/kg) against malathion resistant population of *S. oryzae* compared to laboratory population. In our research, comparison of nominal values of spinosad efficacy against field populations from Serbia with values for laboratory population, showed significant differences only for population from Šid. For example, after 14-days of exposure in wheat treated with 0.25 and 1.0 mg/kg of spinosad, efficacy was 2% and 77%, and progeny reduction 1.3% and 55%, while for laboratory population efficacy was 18% and 96%, and progeny reduction 28% and 81%. However, after application of abamectin, no statistically significant differences were observed in efficacy against tested populations.

With an aim to review total potential of certain insecticide, Subramanyam and Roesli (2000) emphasize the importance and necessity for determination of their indirect effects, where effect on progeny production of storage insects is featured. Having this in mind, we highlight the results on spinosad obtained by Fang et al. (2002) and Nayak et al. (2005), because these results show that this insecticide at rate of 1 mg AI/kg achieves total progeny reduction (100%), but only in *R. dominica*, while the highest progeny reduction in *S. oryzae* amounts 48-54%. In the paper in all weevil populations, spinosad applied at 1 mg/kg reduced progeny by 55-93%. However, Kavallieratos et al. (2009) in wheat treated with 0.1, 0.5 and 1.0 mg/kg of abamectin recorded 100% progeny reduction in *S. oryzae*, *R. dominica* and *T. confusum*, while in our experiments, taking into account all populations and rates of abamectin, high progeny reduction (≥95%) was observed only after *S. oryzae* contact with wheat treated with 1 and 2 mg/kg of abamectin.

How important it is to review susceptibility of storage insects populations and determine appropriate insecticide application rate, for insecticide efficacy evaluation, show the results obtained by Kljajić and Perić (2010) in investigation of effects of sublethal doses (LD_{20} and LD_{50}) of dichlorvos, malathion, chlorpyrifos-methyl, pirimiphos-methyl and deltamethrin on progeny production in three populations of *Sitophilus granarius* (L.) with different susceptibility to insecticides. It was found that sublethal doses of certain contact insecticides can significantly reduce the number of offspring of surviving *S. granarius* specimens, but also to initiate increase in their number, which is certainly in connection with history of insecticide application in control or forced selection of each population. Therefore, based on the results of our research, it can be concluded that for highly efficient control of different *S. oryzae* populations in wheat grain, it is necessary to apply ≥2 mg/kg of abamectin, and particularly of spinosad.

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Efekti spinosada i abamektina na različite populacije pirinčanog žiška *Sitophilus oryzae* (L.) u tretiranoj pšenici u zrnu

REZIME

U laboratorijskim uslovima (25±1ºC i 60±5% r.v.) su ispitivani efekti prirodnih insekticida spinosada i abamektina na pet populacija *S. oryzae* (laboratorijska, Šid, Gornji Milanovac, Žabari i Novi Pazar). Oba insekticida su za sve testirane populacije naneti na netretiranu pšenice u zrnu u dozama 0,25, 0,5, 1,0 i 2,0 mg a.m./kg, nakon čega je u plastične posude (V=200 cm^3) sa po 50 g tretirane pšenice, u četiri ponavljanja za svaku testiranu populaciju, stavljano po 25 adulta *S. oryzae*. Smrtnost žižaka je utvrđivana posle dva, sedam i 14 dana, a posle ukupno osam nedelja od izlaganja roditelja je utvrđivan uticaj na produkciju potomstva.

Efikasnost spinosada i abamektina je posle dva dana izlaganja žižaka kod svih testiranih populacija i svih količina primene bila <15%. Posle sedam dana izlaganja efikasnost od ≥95% utvrđena kod žižaka iz Žabara, u pšenici tretiranoj dozom od 2 mg/kg spinosada i abamektina, i kod žižaka iz Gornjeg Milanovca, samo u pšenici tretiranoj dozom od 2 mg/kg spinosada. Posle 14 dana izlaganja efikasnost ≥95% je utvrđena kod laboratorijskih žižaka i žižaka iz Žabara i Gornjeg Milanovca, u pšenici tretiranoj dozama od 1 i 2 mg/kg spinosada, i kod *S. oryzae* iz Novog Pazara i Šida, u pšenici tretiranoj dozom od 2 mg/kg spinosada. Istovremeno je kod svih testiranih populacija abamektin u dozama 0,5, 1,0 i 2,0 mg/kg bio efikasan 94-100%. Nijedan insekticid nije potpuno (100%) redukovao potomstvo ispitivanih populacija pirinčanog žiška, dok je visoka redukcija potomstva (≥95%) utvrđena samo u kontaktu žižaka sa pšenicom tretiranom dozama od 1 i 2 mg/kg abamektina. Rezultati istraživanja su pokazali da je za visoko efektivno suzbijanje različitih populacija *S. oryzae* u pšenici u zrnu potrebno primeniti ≥2 mg/kg abamektina, i, posebno, spinosada

Ključne reči: *S. oryzae;* pšenica u zrnu; spinosad; abamektin; efekti