SELECTIVITY OF REGISTERED AGROCHEMICALS FOR SOYBEANS AIMING IMMATURE PHASES OF *TRICHOGRAMMA PRETIOSUM*

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ABSTRACT – The parasitoid *Trichogramma pretiosum* has great potential in controlling caterpillars, promoting a reduction in insecticide costs. However, incorrect application with mixtures with agrochemicals often results in failure and discredit of technology. The objective of this work was to evaluate, under laboratory conditions, the selectivity of pesticides reported for soybean in the immature stages (egg, larvae, pre-pupae and pupae) of parasitoids of *Trichogramma pretiosum*. Bioassays were conducted in the laboratory by exposing eggs parasited, using the methods proposed by the International Organization for Biological and Integrated Control of Noxious Animals and Plants (IOBC). For this, the parasitoids were obtained in the laboratory using Anagasta kuehniella eggs as a host for the multiplication of *T. pretiosum.* Temperature (25 ± 1 ºC), relative humidity (70 ± 10%) and photophase (14 hours) conditions were controlled both for the creation of parasitoids in climatic chambers and during bioassays in test rooms. Sixteen insecticides, six fungicides and seven herbicides registered for soybean cultivation in three immature stages of T. pretiosum were evaluated. The experimental design was completely randomized with eight replicates per treatment. Talcord 250, Nexide and Valon 384 EC are the most harmful to emergence of *T. pretiosum* in the larvae egg stage, while Platinum neo is more harmful in the pupal stage. Fox fungicide presented as the most harmful on the emergence of *T. pretiosum* in the immature stages of egg larva and pre pupa. Herbicides do not affect the emergence of *T. pretiosum* when sprayed on the immature stages of egg larva, pre pupa and pupa.

Keywords: Phytosanitary control, insects management, interactions.

SELETIVIDADE DE AGROQUÍMICOS REGISTRADOS PARA SOJA VISANDO FASES IMATURAS DE *TRICHOGRAMMA PRETIOSUM*

RESUMO – O parasitóide *Trichogramma pretiosum* apresenta grande potencial no controle de lagartas, promovendo redução nos custos com inseticidas. No entanto, a aplicação incorreta com misturas com agroquímicos muitas vezes resulta em fracasso e descrédito da tecnologia. O objetivo deste trabalho foi avaliar, em condições de laboratório, a seletividade de defensivos relatados para soja nos estágios imaturos (ovo, larva, pré-pupa e pupa) de parasitóides de *Trichogramma pretiosum*. Os bioensaios foram conduzidos em laboratório com a exposição de ovos parasitados, utilizando-se os métodos propostos pela Organização Internacional para o Controle Biológico e Integrado de Animais e Plantas Nocivas (IOBC). Para isso, os parasitóides foram obtidos em laboratório utilizando ovos de *Anagasta kuehniella* como hospedeiro para multiplicação de *T. pretiosum*. As condições de temperatura (25 ± 1 ºC), umidade relativa (70 ± 10%) e fotofase (14 horas) foram controladas tanto para a criação de parasitóides em câmaras climáticas quanto durante a realização de bioensaios em salas de testes. Foram avaliados 16 inseticidas, seis fungicidas e sete herbicidas registrados para o cultivo da soja em três estádios imaturos de *T. pretiosum*. O delineamento experimental foi inteiramente casualizado com oito repetições por tratamento. Talcord 250, Nexide e Valon 384 EC são os mais nocivos à emergência de *T. pretiosum* na fase de ovo de larva, enquanto Platinum neo é mais prejudicial na fase de pupa. O fungicida Fox apresentou-se como o mais nocivo na emergência de *T. pretiosum* nos estágios imaturos de ovo larva e pré pupa. Os herbicidas não afetam a emergência de *T. pretiosum* quando pulverizados nos estágios imaturos de larva de ovo, pré-pupa e pupa.

Palavras-chave: Controle fitossanitário, manejo de insetos, interações.

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INTRODUCTION

Soybean crops are subject to the attack of pests since its germination to its harvest. The large diversity of pests that occur in this crop, according to its occurrence and population density, can cause economic loss and, consequently, reduce the yield or product final quality (Degrande & Vivan, 2007). Controlling measures must be taken in order to prevent higher losses throughout techniques that avoid the environmental disequilibrium and guarantee the sustainability in its wide sense.

Among the soybean incident caterpillar complex, the vevlvetbean caterpillar *Anticarsia gemmatalis* Hübner, 1818 (Lepidoptera: Noctuidae) it is the main defoliator, being capable of consume about 110 cm² of leaves (Walker et al., 2000). Other lepidopteries are increasing defoliation, as it is the case of *Pseudoplusia includens* (Lepidoptera: Plusiinae), which has been the most frequent and hard to manage (Bellettini et al., 2008). One of the main strategies of pest population suppression is based on chemicals, being the most used technique against it. In Brazil, the are about 414 different chemicals registered for soybean use, but in its majority with high toxicity, where 181 are registered as insecticides (Agrofit, 2011).

However, as a result of the intensive use of insecticides, a number of negative impacts on the environment have been observed (Goulart et al., 2011), especially with regard to natural enemies and their selectivity to different pesticides. The significant increase in the number of sprayings in the cropping areas, the wide range of action of the applied products and the intensive and indiscriminate sprayings (Hegazi et al., 2007), cause financial losses, unbalance the food chain, raise secondary pests to the category of primary pests and induce selection pressure to resistant pests (Vianna et al., 2011).

Another factor that has been studied by Sosa-Gómez (2005) refers to preventive treatments to the Asian soybean rust. In his work, the author found that the use of fungicides, affects negatively the development and establishment of entomopathogenic fungus as *Nomurae rileyi* that, according to Degrande & Vivan (2007), control caterpillars populations dismissing the use of insecticides in order to control the soybean caterpillar. The presence of parasitoids, predators and pathogens that exert biological control of agriculturally important pest insects is indispensable as a dynamic balance factor in agroecosystems (Landis et al., 2000). These natural controlling agents minimize the need for human intervention in pest control through other insect population reduction methods.

The natural parasitism of *A. gemmatalis* eggs by *Trichogramma pretiosum* in soybean crops was verified by Avanci et al., (2005), Bueno et. al, (2009a); Bueno et al. (2011). Recent studies have sought to evaluate the preference of this egg parasitoid over lepidopteran pests that attack soybean (Milanez et al., 2009; Goulart et al., 2011; Siqueira et al., 2012), highlighting the importance of this parasitoid as an effective biological agent in regulating pest insect populations.

The biological control of pests, especially pest insects of the Order Lepidoptera with the use of *Trichogramma*, aims to reduce the use of insecticides, minimizing their environmental impact. However, the use of insecticides, especially in large areas of cultivation, is necessary when there are high incidences of insects/pests. However, emerging techniques based on biological control are also alternatives that enhance insect/pest control and should be considered within production units. At the same time, the correct use of biological insecticides can keep insect pest populations at low levels, which avoids the use of synthetic insecticides or reduces their use.

The integration of biological and chemical control therefore needs to be validated through selectivity testing, allowing the preservation of the beneficial insect community in the production system. To this end, the working group of the International Organization for Biological Control and Integrated Control of Noxious Animals and Plants (IOBC) established guidelines for conducting bioassays at different stages and categorizing pesticides for selectivity: in the laboratory (phase adult and immature); laboratory / greenhouse (persistence and field) with adult parasitoids as recommended by Hassan et al. (2000) and Hassan & Abdelgader (2001), since the adult phase is the most sensitive to the toxic action of phytosanitary products.

For research involving parasitoids, the *Trichogramma* genre has been selected as an indicator for the Hymenoptera order, especially for their greater susceptibility, and due to its ease of procreation (Hassan, 1998). Thus, *T. pretiosum* was chosen for use in selectivity tests, as it is associated with soybean cultivation as an important natural enemy of lepidopteran pests (Bueno et. al 2009a; Bueno et al., 2011).

Some selectivity studies on the immature phase of *T. pretiosum* have already been performed in soybean, however, they used methodologies that differ from those proposed by IOBC (Cañete, 2005; Bueno et al., 2008; Carmo et al., 2009 and 2010), which did not represent similar conditions of the reality of field applications, because the evaluation was performed from the immersion of parasitized eggs in solutions containing the pesticides.

In this manner, it is possible to see that this area needs further studies and results that follow the general protocol of the IOBC tests, which first calls for the assessment of the effects on the adult parasitoid to obtain selectivity classes in initial toxicity tests to the adult (which was performed in Chapter 1 of this work) and proceed to the sequence of laboratory bioassays with all immature parasitoid phases, phase less sensitive to toxic action of pesticides.

Understanding the toxic action of pesticides on the immature stages of the parasitoid can contribute to the correct use and maximize the efficiency of insect/pest control. This directly reflects on sustainable productions that guarantee the preservation of natural resources, as well as increase food security. In this context, the objective of this work was to evaluate, under laboratory conditions, the selectivity of pesticides reported for soybean in the immature stages (egg, larvae, pre-pupae and pupae) of parasitoids of *Trichogramma pretiosum*.

MATERIALS AND METHODS

The assay was carried out in the Laboratory of Integrated Pest Management of the Federal University of Pelotas (LabMIP-UFPel), located in the city of Pelotas, RS, Brazil. The bioassays selectivity for immature stages were based on the method proposed by the International Organization for Biological and Integrated Control of Noxious Animals and Plants (IOBC) (Hassan et al., 2000; Hassan and Abdelgader, 2001). For this purpose, the parasitoids were obtained from a laboratory using *Anagasta kuehniella* eggs (Zeller, 1879) (Lepidoptera:Pyralidae) as host for *T. pretiosum* multiplication (Parra, 1997). Temperature (25 ± 1 ºC), relative humidity (70 ± 10%) and photophase (14 hours) conditions were controlled both for parasitoid rearing in climate chambers (BOD) and during conduction of bioassays in test rooms.

Pesticides evaluated

It was evaluated sixteen insecticides, 6 fungicides and 7 herbicides registered for soybean cultivation. The percentage of adults' emergence in each treatment was calculated, and the average result used for comparison with the control treatment of each immature phase of *T. pretiosum* in each bioassay, in order to classify the pesticides according to the selectivity, based on the reduction of the emergence percentage. According to the IOBC methodology, pesticides were then classified as harmless (class 1, less than 30% reduction in emergence), mildly harmful (class 2, 30-79%), moderately harmful (class 3, 80-99 %) or harmful (class 4, more than 99%). Identifications as commercial product name, active ingredient, chemical group, maximum tested commercial dosage, concentration of active ingredient and commercial product in the solution are presented in Tab. 1.

Table 1 - Pesticides (insecticides, fungicides and herbicides) evaluated in the selectivity tests for the immature phases of *Trichogramma pretiosum* using maximum dosage of the commercial product registered for soybean crop

Commercial Product	Active ingredient	Chemical group	DC [1]	Company [2]	Cpc [3]
		Insecticides			
Nortox Cypermethrin	cypermethrin	Pyrethroid	0.200	0.025	0.1000
Connect ®	beta-cyfluthrin + imidaclopride	Pyrethroid + Neonicotinoid	0.750	0.0046 + 0.0375	0.3750
Full Engeo	thiametoxam + lambda- cyhalothrin	Neonicotinoid + Pyrethroid	0.200	0.0141 + 0.0106	0.1000
Fastac 100 SC	the lfacipermetrina	Pyrethroid	0.120	0.0060	0.0600
Karate Zeon 250 CS	lambda- cyhalothrin	Pyrethroid	0.015	0.2000	0.0187
Lannate BR	methomyl	Organophosphate	1.000	0.1075	0.5000
Lorsban 480 BR	cloripyrifos	Organophosphate	1.000	0.2400	0.5000
Malathion 500 CE Sultox	malationa	Organophosphate	3.000	0.7500	1.5000
Nexide	gama - cyhalothrin	Pyrethroid	0.250	0.0145	0.1250
Oberon	spiromesifen	Ketoenol	0.600	0.087	0.3000

Continua...

Table 1 - Cont.

Commercial Product	Active ingredient	Chemical group	DC [1]	Company [2]	Cpc [3]
		Insecticides			
Platinum	cypermethrin + thiametoxam	Pyrethroid + Neonicotinoid	0.250	0.1375 + 0.2750	0.1250
Tracer	espinosade	Spinosine	0.050	0.0120	0.0250
Talcord 250	permethrin	Pyrethroid	0.120	0.0150	0.0600
Turbo	beta- cyfluthrin	Pyrethroid	0.200	0.0050	0.1000
Valon 384 SC	permethrin	Pyrethroid	0.065		0.0325
		Fungicides			
Envoy	epoxiconazole + piraclostrobin	Triazole + Strobilurin	0.700	0.0175 + 0.0175	0.3500
Folicur 200 EC	Tebuconazole	Triazole	0.750	0.0750	0.3750
Fox	protioconazole + trifloxystrobin	Triazole + E strobilurin	0.400	0.0350 + 0.0300	0.2000
Kumulus DF	Súlfur	Inorganic	2.500	1.0000	1.200
Native	trifloxystrobin + tebuconazole	Strobilurin + Triazole	0.600	0.0300 + 0.0600	0.3000
Sphere Max	cyproconazole + trifloxystrobin	Triazole + Strobilurin	0.200	0.0160 + 0.0375	0.1000
		Herbicides			
Basagran 600	bentazone	Benzothiadiazinone	1.200	0.3600	0.6000
Dual gold	s - metachlor	Chloroacetanilide	2.000	0.0400	1.0000
Finale	glufosinate - ammonium salt	Substituted homoalanine	4.000	0.3000	2.0000
Gramocil	diuron + dichloride of paraquat	Urea + Bipyridylum	2.000	0.2000 + 0.1000	1.0000
Gramoxone 200	dichloride of paraquat	Bipyridylum	3.000	0.3000	1.5000
Original Roundup	glyphosate	Substituted glycine	5.000	0.9000	2.5000
Zapp Qi	potassium glyphosate salt	Substituted glycine	3.500	0.8750	1.7500

[1] DC = Field dosage (L or kg ha $^{-1}$ of the commercial product) considering a syrup volume of 200 L ha $^{-1}$; [2] C.ia = Concentration (%) of the active ingredient in the mixture used in the bioassays; [3] C.pc = Concentration (%) of commercial product in the mixture used in the bioassays .

Parasitism of eggs of the alternative host *Anagasta kuehniella*

To obtain the parasitoid in the different immature phases, a cardboard card containing *A. kuehniella* eggs was placed in glass cylinders (25cm long x 10cm in diameter) containing *T. pretiosum* adults. Each card had 60 circles of 1cm in diameter, made with 400 ± 50 *A. kuehniella* eggs each. After parasitism, the adults were discarded and the cards placed in climatized Biochemical Oxygen Demand (BOD) chambers to provide adequate conditions for the development of *T. pretiosum*. This procedure was repeated three times at 24 hours (1 day), 72 hours (3 days) and 168 hours (7 days) before spraying, so that parasitoids were obtained in their immature egg-larvae, pre-pupa phases and pupa, respectively, inside the host egg (Cósoli et al.,1999). Thus, it was possible to compose a bioassay, which consisted of test pesticides and three immature phases of *T. pretiosum*. Thus, from each card containing 60 circles, eight circles were selected for each treatment in each phase of parasitoid development.

Statistical analysis

The complementary statistical analysis was performed using the SAS statistical software - Statistical Analysis System (Sas Learning Edition, 2002). The results, regarding the number of parasitized eggs per female, were submitted to the normality test by the " Bartlett's test for equal variances" using the Univariate procedure. If this assumption was not met, nonparametric analysis was performed by the NPar1Way procedure (Kruskal-Walis) and, after proving the existence of differences between treatments, the data were transformed by the

Rank procedure. The comparison of means was obtained by the Bonferroni-Dunn *t* test using the Glm procedure at 5% probability of error. In case of normality of data, the averages were compared by Tukey test (balanced data) and Tukey-Kramer (unbalanced data) using the Glm procedure at 5% probability of error.

RESULTS AND DISCUSSION

Insecticides

In bioassay I (Tab. 2), it was observed that the treatment x stage interaction was not significant (F = 3.960; d.f. = 39; p <0.05) for the adult emergence variable of *T. pretiosum*. There were no significant differences between the developmental stages evaluated. In the egg larva phase, most of the products tested showed a small reduction in emergence, and the Oberon product had no reduction in *T. pretiosum* emergence.

Fastac 100 SC, Turbo and Platinum products produced reductions of 17.70, 19.44, and 19.92%, respectively. In the pre-pupil stage, there were also no significant differences in the treatment-stage interaction (F = 0.81; d.f. = 39; p <0.05). Based on the effect of emergence reductions, insecticides did not show significant effects on the parasitoid at this stage, with reductions ranging from 0 to 2.71% (Tab. 2, bioassay I).

Table 2 - Emergence (% ± EP) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides registered for soybean culture, containing the parasitoid, at different immature stages of development (temperature 25 ± 1ºC; relative humidity 70 ± 10 %; photophase 14 hours).

Treatments	Active ingredient	DC [1]	Emergency (% ± SE)							
			Larva Egg			Pre-pupa			Pupa		
Bioassay I										
Witness	-	-	107.90	±	0.66 aA	110.90	±	6.86 aA	105.30	±	0.29 aA
Fastac	the LFA - cypermethrin	0.120	88.80	±	11.31 aA	107.90	±	4.59 aA	91.30	±	5.269 aA
Oberon	spiromesifen	0.600	111.80	±	6.54 aA	110.00	±	16.79 aA	99.41	±	3.67 aA
Platinum	thiametoxam + cypermethrin	0.250	86.41	±	2.74 aA	109.30	±	5.78 aA	108.90	±	18.18 aA
Turbo	beta-cyfluthrin	0.200	86.92	±	10.79 aA	127.80	±	6.10 aA	82.87	±	3.62 aA
Bioassay II										
Witness	-	-	102.10	±	4.38 aA	101.10	±	2.29 aA	99.60	±	1.15 aA
Curyon 550 EC	lufenuron + profenofós	0.150	100.20	±	5.70 aA	103.70	±	4.21 aA	92.83	±	9.47 aA
Full Engeo	thiametoxam + lambda-cyhalothrin	0.200	94.86	±	3.46 aA	94.51	±	2.67 aA	94.55	±	7.17 aA
Lannate BR	methomyl	1.000	105.10	±	2.44 aA	91.13	±	3.80 aA	91.64	±	2.03 aA
Malathion 500 CE Sultox	malationa	3.000	108.90	±	4.41 aA	85.71	±	3.45 aB	89.52	±	2.66 aA

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

Means followed by the same lowercase letter in the column and uppercase in the row do not differ statistically from Tukey, at the 5% probability level.

In the pupal stage, the stage x treatment interaction (F = 1.49; df = 39; p <0.05), while not being significant, product Oberon had one small reduction in emergence 5.59% at this stage, the largest of the immature phases for the product. The same effect was observed for the Turbo product that had potentiated its action on the parasitoid pupa phase, presenting an emergence reduction of 21.30%. The Platinum product was totally without effect on the pupal phase, presenting a superior emergence including the control, a fact that can be explained by the possibility of developing more than one parasitoid per host egg. Therefore, it was possible to verify that in bioassay I, all tested products were innocuous to the immature phases of *T. pretiosum* (Tab.2).

Stefanello Júnior (2010) working with immature stages of *T. pretiosum* in maize culture observed that for the pyrethroid insecticide Turbo, the pupa phase was the most affected by the insecticide action corroborating the results contained in it and also conferred class 1, for this insecticide. Santos et al. (2016) also revealed greater aggressiveness of beta-cyfluthrin against *T. pretiosum*.

Insecticide Fastac 100 SC, is a pyrethroid insecticide, and at the concentration tested (0.0060) was considered innocuous, causing a slightly more pronounced effect for the egg-larva stage in the present work. According to Bueno et al., (2008), studying soybeans evaluated the same active principle and found different immature classes when testing the dosage of 100 ml.ha where found in the egg and larval stage class 2, which differ from the present work. However, it was used a different methodology of the present work, soaking the parasitized eggs for 5 seconds, immersing the eggs totally in the insecticide formulation. However, the same author corroborating the result of this work, found class 1 for the pupa stage.

Platinum insecticide is derived from a mixture between the pyrethroid + neonicotinoid chemical groups, and has been shown to have minor interference mainly on the egg-larva stage. As it is a relatively new commercial product, no data related to its selectivity were found in the literature. Although not carried selectivity tests with commercial products containing only the active ingredient from the group of neonicotinoids, Moura et al. (2005) found that thiamethoxam (0.05 gL^{-1} of going). At the maximum registered dose for tomato, was innocuous to the immature phases of *T. pretiosum*. Thus, the toxicity attributed to the mixed products of more neonicotinoid pyrethroids may be due to the interaction between both active ingredients, inert ingredients and / or product formulation.

In bioassay II (Tab. 2), it was observed that the treatment x stage interaction was not significant (F = 1. 544; d.f. = 39; p <0.05), as well as no significant differences between the developmental stages evaluated (F = 3.287; d.f. = 14; p <0.05). In the egg- larvae phase Curyom 550 Ec and Engeo Pleno showed a small reduction in emergence, and the products Lannate BR and Malathion 500 CE Sultox did not interfere in the emergence of *T. pretiosum*, showing no reduction in emergence of parasite ide. The full Engeo insecticide showed a 7.09% reduction in emergence while Curyon 550 EC showed a 1.86% reduction.

In the pre-pupil stage, the treatment x stage interaction presents a significant difference (F = 3.936; d.f. = 39; p <0.05) of the Malathion 500 CE Sultox product between the different stages. Despite the statistical difference, no change was observed in the insecticide class, which presented a reduction of 15.22%, being considered innocuous. However, on pre-pupal stage was observed its most dramatic effect on the reduction in the emergence of adult *T. pretiosum*. In the parasitoid pupal stage, treatment x stage interaction was not significant (F = 0.9642; d.f. = 39; p <0.05), as there were no significant differences between the developmental stages evaluated (F = 3.287; d.f. = 39; p <0.05). However, the effect of Malathion 500 EC Sultox in this stage caused further reduction of this insecticide in the stage evaluated.

In the bioassay II, the inoculation of Curyom 550 Ec, Engeo Plenum, Lannate BR and Malathion 500 EC Sultox in immature stages of *T. pretiosum*.

The insecticide Malathion 500 CE Sultox showed a higher interference in the pre-pupa stage, which was observed by other authors. Manzoni et al. (2007) evaluating the active ingredient Malatione using the commercial product Malathion 1000 CE (0.1% ai. in formulation), was classified as innocuous for the egg-larva and pupa phase, although slightly harmful (class 2) to the pre-pupa phase. Nörnberg (2008) classified Malathion 1000 CE (0.1% ai in formulation) as innocuous for the egg-larva and pupa phase and slightly harmful (class 2) for *T. pretiosum* pre-pupa. Stefanello Júnior (2010), testing Malathion 500 CE Sultox in maize crop (0.625% ai in formulation) found that it was innocuous for all immature stages of the parasitoid corroborating the results of this work and revealing that there are greater effects of insecticide in the pre- pupa phase of the parasitoid in question.

The insecticide Lannate BR presented low mortality, with emergence results close to the values found for the control in the present study for *T. pretiosum* (Tab. 3), which differs slightly from the results found in the literature. Hohmann (1991) found that methomyl (0.001075% ai in the formulation) was highly toxic to the immature stages of *T. pretiosum*, which only detected emergence of the parasitoid when it received insecticide treatment after 7 days of development of *A. kuehniella* egg. For Hassan et al. (1987), Lannate (0.1% commercial product in spraying formulation) was harmful (class 4) to the *T. cacoeciae* pupa phase. Bueno et al. (2008), using other methodology and greater field strength (1.5 $l.ha^{-1}$) found product to belong to class 3 for egg-larvae stages and class 2 for the stage of *T. pretiosum* pupa. Stefanello Júnior (2010) found similar results to this work in corn culture bioassays, however using a higher active ingredient concentration in the formulation (0.3%).

Engeo Pleno insecticide showed very similar reductions in the three different stages, with a tendency of higher lethality to egg-larva phase of 7.09 %. However, at the respective concentration (0.0141 + 0.0106) of the thiametoxam + lambda- cyhalothrin mixture showed no significant effects on the emergence of the parasitoids. Stefanello Júnior (2010) observed that Engeo Pleno was innocuous (class 1) for egg-larva and slightly harmful (class 2) for pre-pupa and pupa, differing from the present study where the insecticide was innocuous, being the possible explanation for this. In fact, the highest concentration of active ingredient contained in the formulation as a function of the higher field dosage recorded in corn crop.

In bioassay III, it was observed that the treatment x stage interaction was not significant (F = 2.407; d.f. = 47; p <0.05) for the adult emergence variable of *T. pretiosum* (Tab. 3). No significant differences were observed between the developmental stages evaluated (F = 2.284; d.f. = 17; p <0.05). In the larval egg stage Connect, Karate Zeon 250 CS and Tracer insecticides caused reductions ranging from 4.66 to 8.68% on emergence of the parasitoid. However, Talcord 250 and Lorsban 480 BR, despite not presenting statistical differences, were quite aggressive to the egg-larva stage, causing reductions in emergence of 27.25 and 20.25%, revealing a high toxicity to this stage.

Table 3 - Emergence (% ± EP) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides registered for soybean culture, containing the parasitoid, at different immature stages of development (temperature 25 ± 1ºC; relative humidity 70 ± 10 %; photophase 14 hours).

Treatments	**Active ingredient**	**DC [1]**	**Emergency (% ± SE) [2]**								
			Larva Egg			**Pre-pupa**			**Pupa**		
Bioassay III											
Witness	-	-	105.10	±	3.03 a A	102.30	±	3.61 aA	109.70	±	3.12 aA
Connect	beta - cyfluthrin + imidacloprid	0.150	100.20	±	2.56 aA	95.73	±	8.78 aA	99.18	±	1.55 aA
Lorsban 480 BR	chloripyrifos	1.000	83.82	±	11.31 aA	108.30	±	2.22 aA	81.85	±	12.64 aA
Talcord 250	permethrin	0.120	76.46	±	10.78 aA	94.57	±	16.29 aA	114.1	±	4.13 aB
Tracer	espinosade	0.050	93.08	±	4.01 aA	119.40	±	6.00 aA	95.98	±	2.74 aA
Zeon Karate 250	lambda-cyhalothrin	0.030	95.98	±	3.92 aA	106.10	±	4.30 aA	105.80	±	3.39 aA
Bioassay IV											
Witness	-	-	109.03	±	2.63 aA	105.90	±	0.79 aA	95.80	±	3.47 aA
Nexide	gama - cyhalothrin	0.250	79.48	±	0.92 aC	85.90	±	4.20 aB	84.35	±	3.02 aA
Valon 384 EC	permethrin	0.065	79.16	±	0.87 aC	98.07	±	1.03 aA	82.51	±	3.05 aA
Nortox Cypermethrin	cypermethrin	0.200	85.64	±	1.69 aB	99.32	±	0.87 aA	89.27	±	3.08 aA
Platinum neo	lambda-cyhalothrin + thiametoxam	0.200	80.10	±	1.06 aB	82.89	±	1.23 aB	74.16	±	3.55 aB

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

Means followed by the same lowercase letter in the column and uppercase in the row do not differ statistically from Tukey, at the 5% probability level.

According to the selectivity classes assigned to insecticides, Connect, Iorsban 480 Br, Talcord 250, Tracer and Karate Zeon 250 CS were innocuous (class 1) to the pre-pupal egg-larva and pupa phases of *T. pretiosum*.

For the insecticide Karate Hassan (1994 a) rated it as slightly harmful to the pupation of *Trichogramma cacoeciae* Marchal, 1927 Hymenoptera: Trichogrammatidae).

According to the emergence reductions observed for the insecticides of the pyrethroid chemical group, containing only the active ingredient in this group, most were considered innocuous for all immature stages of *T. pretiosum* development.

For the insecticide Lorsban 480 BR (0.00072% a.i. in mixture), Giolo (2007) and Nörnberg (2008) obtained class 2 (slightly harmful) for all immature phases of *T. pretiosum*, unlike that obtained in the present study, where the product is harmless (class 1) for all immature stages. However & Maia (2009) found that the active ingredient present in the commercial product Astro (0.005% commercial product in the formulation) was innocuous to the immature phases of *T. atopovirilia*, although in a different concentration to the present work, which used Lorsban 480 BR. (0.24% commercial product in the formulation).

Bueno et al., 2008 testing Lorsban 240 in soybean crop using the immersion methodology conferred class 1 for the egg phase, similar to the present study. However, they considered it to be slightly harmful to the larval and pupal stages of the parasitoid. Stefanello Júnior (2010) considered that the same product was moderately harmful (class 3) for the egg-larva and pre-pupa phases and harmful (class 4) for the *T. pretiosum* pupa phase, however, using the dosage 0.5% commercial product in the syrup. However, it was possible to verify that the reduction was higher in the pupa phase for the active ingredient concentration tested (0.24%).

For the insecticide Tracer (0.00006% a.i. in formulation) Giolo (2007) classified it as innocuous for all immature phases of the parasitoid, similar to the present study (0.01%). Tracer (0.1% of the commercial product in the formulation) was considered slightly harmful to *T. atopovirilia* by Maia (2009) when evaluating the product in corn crop. Stefanello Júnior (2010), with a concentration of 0.05% of commercial product in the formulation, obtained class 4 for the same insecticide. Carmo et al. (2010), working with the soybean culture on the pupal phase considered that Tracer (24 g a.i. ha^{-1}) was classified as moderately harmful (class 3) because it reduced the emergence of the parasitoids of 82.24% in relation to the control treatment. Similar results with this product at the dose of 12 g a.i. ha^{-1} were obtained by Cañete (2005) for five different *Trichogramma* species, as well as Bueno et al. (2008), whom obtained class 4 for all immature parasitoid phases. It is possible to verify that the dosage of the mentioned works are 240 and 480 times higher than the tested dose, what explains the difference between the obtained classes, besides using methodologies that differ from the present study, which excels in the standardized methodology proposed by IOBC.

The Connect insecticide, which presented sensitive effects to the 3 stages of parasitoid development was assigned class 1 (innocuous) with reductions in parasitism of 4.66, 6.42, and 9.59% (Tab. 3). Carmo et al. (2010) classified the same product as slightly harmful (class 2) to the pupae phase of *T. pretiosum*. Similar results were obtained by Matos (2007) who observed no harmful effect of imidaclopride on the pupae phase of *T. pretiosum*. The same pesticide dosage when applied to pupae of *Telenomus remus* Nixon, 1937 (Hymenotpera : Scelionidae) was classified as selective (Carmo et al., 2009) which was even more tolerant of the action of this insecticide than *T. pretiosum* . Thus, this product has been shown to be compatible for use in IPM because it preserves the action of the parasitoid *T. pretiosum* in its immature phases, for programmed use in a management program, since its action on adults is harmful.

In the IV bioassay, it was observed that the treatment x stage interaction was significant ($F = 66.36$; d.f. = 39; $p < 0.05$) for all 3 stages of parasitoid, egg-larva, pre- pupa and pupa, respectively (Tab. 3). However, statistically there were no significant differences between the developmental stages evaluated ($F = 1.123$; d.f. = 14; $p < 0.05$).

The insecticide cypermethrin Nortox, Valon 384 EC, Nexide and Platinum C caused considerable reductions in egg-larvae stage, being more pronounced among all the other stages, as can be checked in the bioassay IR (Tab. 3).

In the pre- stage Platinum Neo and Nexide differed from the control reaching reductions of 21.30% and 18.89%, being products that should be observed with caution because they present high rates of reduction in parasitism in this stage.

In the pupal stage, the largest reduction was found for the insecticide Platinum Neo, which showed a reduction in the emergence of *T. pretiosum* adults 22.59% and differed from the other treatments within this stage.

According to the IOBC ratings given for the evaluated insecticides, all products were innocuous (class 1) for all *T. pretiosum* immature phases (Tab. 4 and 5). Among the immature phases of *T. pretiosum*, the pupa phase was not always considered the most tolerant to the effects of pesticides according to IOBC assumptions (Hassan et al., 2000; Hassan & Abdelgader, 2001).

Table 4 - Reduction in emergence (%) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides recorded for soybean-containing parasitoid at different immature stages of development and selectivity classification according to IOBC / WPRS (temperature 25 ± 1ºC; relative humidity 70 ± 10%; photophase 14 hours).

Treatments	Active ingredient	DC [1]	Immature phase					
			Larva Egg		Pre-pupa		Pupa	
			RE [2]	C[3]	RE	C[3]	RE	C[3]
Bioassay I								
Fastac	alphaacipermethrin	0.120	17.70	1	2.71	1	13.30	1
Oberon	spiromesifen	0.600	0.00	1	0.81	1	5.59	1
Platinum	thiametoxam + cypermethrin	0.250	19.92	1	1.44	1	0.00	1
Turbo	beta-cyfluthrin	0.200	19.44	1	0.00	1	21.30	1
Bioassay II								
Full Engeo	thiametoxam + lambda-cyhalothrin	0.10	7.09	1	6.52	1	5.07	1
Lannate BR	methomyl	0.08	0.00	1	9.86	1	7.99	1
Malathion 500 CE	malationa	0.04	0.00	1	15.22	1	10.12	1
Curyon 550 EC	lufenuron + profenofós	0.10	1.86	1	0.00	1	6.80	1

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

RE =% reduction in emergency

C[3] = IOBC Classes: 1-Harmless (<30%), 2 = Slightly Harmful (30-79%) 3 = Moderately Harmful (80-99%), 4 = Harmful (> 99%).

Table 5 - Reduction in emergence (%) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides recorded for soybean containing parasitoid at different immature stages of development and classification of selectivity according to IOBC / WPRS (temperature 25 ± 1ºC; relative humidity 70 ± 10%; photophase 14 hours).

Treatments	Active ingredient	DC [1]	Immature phase					
			Larva Egg		Pre-pupa		Pupa	
			RE [2]	C[3]	RE	C[3]	RE	C[3]
Bioassay III								
Connect	beta-cyfluthrin + imidaclopride	0.150	4.66	1	6.42	1	9.59	1
Lorsban 480 BR	chloripyrifos	0.025	20.25	1	0.00	1	25.39	1
Talcord 250	permethrin	0.06	27.25	1	7.56	1	0.00	1
Tracer	espinosade	0.065	11.44	1	0.00	1	12.51	1
Zeon Karate 250	lambda-cyhalothrin	0.10	8.68	1	0.00	1	3.56	1
Bioassay IV								
Valon 384 EC	permethrin	0.10	27.40	1	7.39	1	13.87	1
Nortox Cypermethrin	cypermethrin	0.08	21.45	1	6.21	1	6.82	1
Platinum Neo	lambda-cyhalothrin + thiametoxam	0.04	25.53	1	21.73	1	22.59	1
Nexide	gamma cyhalothrin	0.10	27.10	1	18.89	1	11.95	1

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

RE =% reduction in emergency

C[3] = IOBC Classes: 1-Harmless (<30%), 2 = Slightly Harmful (30-79%) 3 = Moderately Harmful (80-99%), 4 = Harmful (> 99%).

Fungicides

Results of fungicides effects on the emergence of *T. pretiosum* are shown in Tab. 6 and those regarding the selectivity classification, according to IOBC / WPRS, are expressed in Tab. 7. In bioassay V (Tab. 4), the treatment-stage interaction was not significant (F = 0.7663; d.f. = 8; p = <0.06084) for the emergence of *T. pretiosum* adults.

Among the fungicides, they differed only within the stages in relation to the immature developmental stages of *T. pretiosum*, Fox and Folicur 200 EC showed reductions in emergence (Tab. 6), but did not interfere in the class fixed by the IOBC parameters, and precisely in the egg-larva phase. (Tab. 7).

Table 6 - Emergence (% ± EP) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides registered for soybean culture, containing the parasitoid, at different immature developmental stages (temperature 25 ± 1ºC; relative humidity 70 ± 10 %; photophase 14 hours)

Treatments	Active ingredient	DC [1]	Emergence (% ± SE) [2]								
			Larva Egg			Pre-pupa			Pupa		
Bioassay V											
Witness	-	-	107.01	±	9.73 ab A	102.24	±	4.97 ab A	103.76	±	5.88 a A
Envoy	epoxiconazole + p iraclostrobin	0.700	100.92	±	1.98 a A	100.44	±	7.30 ab A	101.11	±	4.16 a A
Follicle	tebuconazole	0.750	96.06	±	25.16 ab A	114.83	±	17.15 a A	101.92	±	8.79 a A
Fox	protioconazole + trifloxystrobin	0.400	93.01	±	10.02 B A	97.99	±	8.83 b A	101.67	±	3.47 a A
Kumulus DF	sulfur	2.500	104.68	±	9.83 ab A	105.51	±	35.43 ab A	101.56	±	3.12 a A
Native	trifloxystrobin + t ebuconazole	0.600	100.41	±	6.42 ab A	106.96	±	17.96 ab A	101.93	±	3.59 a A
Sphere max	cyproconazole + trifloxystrobin	0.200	102.30	±	10.89 ab A	106.66	±	3.74 ab A	103.48	±	4.89 a A

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

Means followed by the same lowercase letter in the column and uppercase in the row do not differ statistically from Tukey, at the 5% probability level.

Table 7 - Reduction in emergence (%) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with fungicides registered for soybean-containing parasitoid at different immature stages of development and selectivity classification according to IOBC / WPRS (temperature 25 ± 1ºC; relative humidity 70 ± 10%; photophase 14 hours)

Treatments	Active ingredient	DC [1]	Immature phase					
			Larva Egg		Pre-pupa		Pupa	
			RE [2]	C[3]	RE	C[3]	RE	C[3]
Bioassay V								
Envoy	epoxiconazole + p iraclostrobin	0.700		1	1.76	1	2.55	1
Follicle	tebuconazole	0.750	10.23	1	0.00	1	1.77	1
Fox	protioconazole + trifloxystrobin	0.400	13.08	1	4.15	1	2.01	1
Kumulus DF	sulfur	2.500	2.18	1	0.00	1	2.12	1
Native	trifloxystrobin + tebuconazole	0.600	6.17	1	0.00	1	1.76	1
Sphere max	cyproconazole + trifloxystrobin	0.200	4.40	1	0.00	1	0.99	1

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

RE[2] =% reduction in emergency

C[3] = IOBC Classes: 1-Harmless (<30%), 2 = Slightly harmful (30-79%) 3 = Moderately harmful (80-99%), 4 = Harmful (> 99%);

The pre-pupa phase was the least affected by the action of fungicides. The pupal phase, however, tended to be more affected when we observed the reduction in the emergence of *T. pretiosum* adults in relation to the other stages, as it suffered minor influences, however from all tested products.

The Fox product, on the other hand, caused small reductions in the emergence rate, affecting the three stages of development, but incurring small reductions that did not influence its harmlessness to the immature stages of this natural enemy. Although small reductions in emergence were observed, it was considered the fungicide with the greatest reduction in emergence of *Trichogramma pretiosum.* Khan et al. (2017) revealed similar results, which did not observe a significant action of fungicides on the emergence of *Trichogramma pretiosum*.

On the other hand, Sphere Max and Envoy fungicides showed emergence reduction rates only for the egg-larva and pupa phases, with emergence averages higher than the control in the pre-pupa phase. Both fungicides were classified as harmless to the immature stages of *T.pretiosum.*

Despite being classified in class 4, when exposed to adults, the Kumulus DF product presented little reduction in emergence rates in spite of the high dosage applied (2.5 kg / ha), and it was conferred to class 1. Grützmacher et al. (2004 a), when evaluating the toxicity of Kumulus DF to the immature phases of *T. cacoeciae,* found the harmlessness of the same product, corroborating the results obtained by Giolo et al. (2006) on *T. pretiosum*, for peach culture and by Manzoni et al. (2007) for the apple crop.

Folicur 200 EC Fungicide was considered class 1 selective for the maximum field dosage in soybean crop. Similar results were found for Folicur 250 EC (0.0625% active ingredient in syrup) by Sterk et al. (1999), who classified the fungicide as innocuous for the *T. cacoeciae* pupa phase.

Herbicides

In bioassay VI (Tab. 8), the emergence variable of *T. pretiosum* differed significantly for the treatment x stage interaction (F=4.95; d.f.=8; p= <.0001). Among the herbicides, for the egg-larva phase, the reduction in emergence was found only on Basagran 600 and Gramocil, and the other treatments presented higher averages than the control treatment. In the pre-pupa phase, only the Dual gold product maintained its average higher than the control treatment (Tab. 8). In the pupal stage, all products had effects on the parasitoid emergence, however, all products demonstrated safety to the immature stages of the parasitoid.

Herbicide work has been carried out for some years by the UFPEL Economic Entomology Research Group. Roundup and Finale have had their selectivity studied by Manzoni et al. (2007), in research using dosages recorded for apple, Roundup Original (2.16%) and Finale (0.2%). As in the present work, they were innocuous for the immature phases of *T. pretiosum*. Later the same results were found for both herbicides by Nörnberg (2008) and Stefanello Junior (2010).

Although the selectivity based on the IOBC parameters was found, the herbicide Basagran showed the most considerable effects on all stages of parasitoid development, as it showed reductions in emergence close to the limits established for class 1 (Tab. 9).

For the herbicide Gramoxone 200, the most notable effect was on the pupal phase, however the product showed a 15.45% reduction in emergence, differing somewhat from the results by Stefanello Junior (2008). This author also found greater toxicity due to the greater reduction in emergence of *T. pretiosum* for pupal phase.

This information corroborates the harmlessness of these commercial products for the dosages and natural enemies tested, and allows greater safety regarding their use in soybean crops. It can be verified that the herbicides tested do not affect the emergence of *T. pretiosum* when sprayed on the immature stages of egg larva, pre pupa and pupa, and can be compatible with biological control.

These evidenced results corroborate the evidence of Khan et al. (2017), where they found herbicides that do not exhibit toxic action on an emergence of Trichogramma pretiosum. Therefore, these herbicides represent a potential option for weed control with low toxicity to the parasitoid T. pretiosum.

Table 8 - Emergence (% ± EP) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with insecticides registered for soybean culture, containing the parasitoid, at different immature developmental stages (temperature 25 ± 1ºC; relative humidity 70 ± 10 %; photophase 14 hours).

Treatments	Active ingredient	DC [1]	Emergence (% ± SE) [2]								
			Larva Egg			Pre-pupa			Pupa		
Bioassay VI											
Witness	-	-	96.57	±	3.19 a A	124.00	±	5.81 a A	97.42	±	6.70 a A
Basagran 600	bentazone	1.2	87.70	±	9.24 a A	95.22	±	14.02 a A	73.33	±	9.15 a A
Dual gold	s - metachlor	2.0	97.42	±	5.20 a A	135.7	±	3.28 a A	90.09	±	1.55 a A
Finale	glufosinate - ammonium salt	4.0	100.03	±	5.85 a A	87.97	±	11.92 a B	88.42	±	7.49 a A
Gramocil	diuron + paraquat dichloride	2.0	84.30	±	4.59 a B	108.50	±	13.25 a A	82.36	±	8.50 a A
Gramoxone 200	paraquate dichloride	3.0	105.08	±	3.30 a A	113.50	±	3.52 a A	82.36	±	2.02 a A
Original Roundup	glyphosate	5.0	109.60	±	5.16 a A	118.20	±	3.71 a A	81.29	±	8.50 a A
Zapp Qi	potassium glyphosate salt	3.5	96.79	±	4.02 a A	108.80	±	9.78 a A	88.35	±	2.24 a A

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

Means followed by the same lowercase letter in the column and uppercase in the row do not differ statistically from Tukey, at the 5% probability level.

Table 9 - Reduction in emergence (%) of *Trichogramma pretiosum* when eggs of host *Anagasta kuehniella* were sprayed with soybean-recorded herbicides containing the parasitoid at different immature developmental stages and selectivity classification according to IOBC / WPRS (temperature 25 ± 1ºC; relative humidity 70 ± 10%; photophase 14 hours). Pelotas, RS.

Treatments	Active ingredient	DC [1]	Immature phase					
			Larva Egg		Pre-pupa		Pupa	
			RE[2]	C[3]	RE	C[3]	RE	C[3]
Bioassay VII								
Basagran 600	Bentazone	1.2	9.18	1	23.20	1	24.72	1
Dual gold	s - metachlor	2.0	0.00	1	0.00	1	7.52	1
Finale	glufosinate - ammonium salt	4.0	0.00	1	29.05	1	9.23	1
Gramocil	diuron + paraquat dichloride	2.0	12.70	1	12.50	1	15.45	1
Gramoxone 200	paraquate dichloride	3.0	0.00	1	8.46	1	15.45	1
Original Roundup	Glyphosate	5.0	0.00	1	4.67	1	16.55	1
Zapp Qi	potassium glyphosate salt	3.5	0.00	1	12.25	1	9.31	1

DC[1] = Commercial product dosage (kg or L ha $^{-1}$);

RE[2] =% reduction in emergency

C[3] = IOBC Classes: 1-Harmless (<30%), 2 = Slightly harmful (30-79%) 3 = Moderately harmful (80-99%), 4 = Harmful (> 99%);

CONCLUSIONS

Talcord 250, Nexide and Valon 384 EC are the most harmful to emergence of T. pretiosum in the larvae egg stage, while Platinum neo is more harmful in the pupal stage.

Fox fungicide presented as the most harmful on the emergence of *T. pretiosum* in the immature stages of egg larva and pre pupa.

Herbicides do not affect the emergence of *T. pretiosum* when sprayed on the immature stages of egg larva, prepupa and pupa.

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