Interaction between *Telenomus remus* and *Trichogramma pretiosum* in the management of *Spodoptera* spp.

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ABSTRACT. Interaction betweeen *Telenomus remus* and *Trichogramma pretiosum* in the management of *Spodoptera* spp. The use of egg parasitoids is a promising strategy for Integrated Pest Management (IPM), but different species of parasitoids have greater or lesser control efficiency, depending on the pest species. Recently, not only *Anticarsia gemmatalis* and *Pseudoplusia includens* but also *Spodoptera cosmioides* and *S. eridania* have been among the key Lepidoptera larvae attacking soybeans. This study evaluated the combination of *Telenomus remus* and *Trichogramma pretiosum* for parasitism of eggs of the *Spodoptera* complex, for better control efficiency and broader spectrum of action among the key pests of soybeans. The experiment was carried out under controlled environmental conditions (25 ± 2ºC; 70 ± 10% RH; and 14 h photophase) in a completely randomized experimental design with seven treatments and 10 replicates with *S. frugiperda*, *S. cosmioides* and *S. eridania* eggs. Each replicate consisted of one egg mass of each *Spodoptera* species, with approximately 100 eggs offered to the parasitoids. The treatments were: 1) 10 females of *T. pretiosum*; 2) nine females of *T. pretiosum* and one female of *T. remus*; 3) eight females of *T. pretiosum* and two females of *T. remus*; 4) seven females of *T. pretiosum* and three females of *T. remus*; 5) six females of *T. pretiosum* and four females of *T. remus*; 6) five females of *T. pretiosum* and five females of *T. remus*, and 7) 10 females of *T. remus*. The parameter evaluated was the percentage of parasitized eggs. Results showed that treatments combining both parasitoid species with only 1 *T. remus* for each 9 *T. pretiosum* (10%) and only 2 *T. remus* for each 8 *T. pretiosum* (20%) were enough to significantly increase the parasitism observed on eggs of *S. cosmioides* and *S. frugiperda*, respectively. This association of *T. pretiosum* and *T. remus* in different proportions is very promising for biological control in IPM programs because it provides wide spectrum of control.

KEYWORDS. Biological control; parasitism; Scelionidae; Trichogrammatidae.

RESUMO. Interação de *Telenomus remus* e *Trichogramma pretiosum* no manejo de S*podoptera* spp. O uso de parasitóides de ovos é uma estratégia promissora dentro do manejo integrado de pragas (MIP), mas diferentes espécies de parasitóides têm maior ou menor eficiência dependendo da espécie praga. Recentemente, não apenas *Anticarsia gemmatalis* e *Pseudoplusia includens*, mas também *Spodoptera cosmioides* e *S. eridania* estão entre as principais larvas de Lepidoptera que estão atacando a cultura da soja. Assim, este trabalho avaliou a possibilidade do uso associado de *Telenomus remus* e *Trichogramma pretiosum* no controle de ovos do complexo *Spodoptera*, objetivando uma maior eficiência de parasitismo com um maior espectro de ação entre as pragas-chave desta cultura. O experimento foi conduzido em condições controladas (25 ± 2ºC; 70 ± 10% e fotofase de 14 h) no delineamento inteiramente casualizado com 7 tratamentos e 10 repetições e ovos de *S. frugiperda*, *S. cosmioides* e *S. eridania*. Para cada repetição foi oferecida aos parasitóides uma postura de cada uma das espécies de *Spodoptera*, com aproximadamente 100 ovos. As diferentes proporções dos parasitóides (tratamentos) avaliadas foram: 1) dez fêmeas de *T. pretiosum* 2) nove fêmeas de *T. pretiosum* e uma fêmea de *T. remus* 3) oito fêmeas de *T. pretiosum* e duas fêmeas de *T. remus* 4) sete fêmeas de *T. pretiosum* e três fêmeas de *T. remus* 5) seis fêmeas de *T. pretiosum* e quatro fêmeas de *T. remus* 6) cinco fêmeas de *T. pretiosum* e cinco fêmeas de *T. remus* 7) dez fêmeas de *T. remus*. O parâmetro avaliado foi à porcentagem de ovos parasitados. Os resultados mostraram que tratamentos combinando ambas as espécies de parasitóides com apenas 1 *T. remus* para cada 9 *T. pretiosum* (10%) e apenas 2 *T. remus* para cada 8 *T. pretiosum* (20%) foram suficientes para aumentar significativamente o parasitismo observado em ovos de *S. cosmioides* e *S. frugiperda*, respectivamente. Esta associação de *T. pretiosum* e *T. remus* em diferentes proporções é bastante promissora para utilização em programas de controle biológico dentro do MIP visto que poderá fornecer um bom espectro de controle.

PALAVRAS-CHAVE. Controle biológico; parasitismo; Scelionidae; Trichogrammatidae.

Soybean is one of the most important export products of Brazil, and in the crop season 2008/2009 57.1 million tons were produced in the country (CONAB 2009). Currently, in the main Brazilian production regions, besides the key pest of this crop, the velvetbean caterpillar, *Anticarsia gemmatalis* Hübner, 1818 (Lepidoptera, Noctuidae), other species of Lepidoptera have caused extensive damage on soybean crops. Among them are other Noctuidae, such as the soybean-looper, *Pseudoplusia includens* Walker, 1857 and some species of the genus *Spodoptera*, as the southern armyworm, *Spodoptera eridania* Cramer, *Spodoptera cosmioides* Walker (Bueno *et al.* 2007) and the fall armyworm, *Spodoptera frugiperda* J. E. Smith, 1797.

In order to maximize agricultural production, pest control should have an interdisciplinary and multidisciplinary vision, integrating different control methods that are less harmful to humans and to the environment. In this context, the adoption of additional tactics for the successful control of insect pests can be incorporated within the IPM philosophy. Among those control tactics, parasitoid releases have shown good results, particularly for the management of insect pests of the order Lepidoptera (Parra 2006). The egg parasitoids of the genus *Trichogramma* (Hymenoptera, Trichogrammatidae) have been among the most studied and used species. The use of parasitoids have several advantages, such as easy rearing on alternative hosts and massive releases for the control of key pests of several crops such as cotton, sugar cane, vegetables, corn, soybean, tomato, and also stored grains (Parra & Zucchi 2004).

Despite the generalist feeding habits, the parasitoids of the genus *Trichogramma* are typically less efficient in controlling species of *Spodoptera*. This is more evident in species that oviposit in overlapping layers, since these parasitoids are able to parasitize only the upper layers of the egg masses. In this context, an efficient egg parasitoid is *Telenomus remus* Nixon (Hymenoptera, Scelionidae), because it is notable for parasitizing even those eggs located in the inner layers of the egg masses (Cruz & Figueiredo 1994). Thus, *T. remus* can be considered as one of the species with the greatest potential for the biological control of *Spodoptera* spp. (Johnson 1984). However, there is a barrier to the use of *T. remus* on large scale release, because it cannot be reared on alternative hosts. This important fact would increase costs, since the rearing of *Spodoptera* is more costly and complex than rearing alternative hosts used for *Trichogramma* spp. Thus, the association of *T. remus* with *T. pretiosum* may be a good option for the management of the S*podoptera* complex. Therefore, the objective of this study was to evaluate the association of *T. remus* and *T. pretiosum* to control larvae of the *Spodoptera* complex.

MATERIAL AND METHODS

This research was carried out in laboratory in controlled environmental conditions of 25 ± 2ºC temperature, 70 ± 10% relative humidity, and 14 h photophase. It was used a completely randomized experimental design, with seven treatments and 10 replicates using *S. frugiperda*, *S. cosmioides* and *S. eridania* eggs. The insects for the experiments were from laboratory rearing, and the adults of *T. pretiosum* and *T. remus* were multiplied on eggs of *Anagasta kuehniella* Zeller, 1879 (Lepidoptera, Pyralidae) and *S. frugiperda*, respectively.

Previously defined proportions of *T. remus* and *T. pretiosum* females were placed into 10 cm high x 2 cm in diameter glass vials, containing a droplet of honey for the parasitoids to feed on. The *S. frugiperda*, *S. eridania* and *S. cosmioides* egg masses, with about 100 eggs each, were glued to 4 cm x 1.5 cm white cardboard and introduced in the tubes containing the different proportions of each parasitoid. The treatments were: 1) 10 females of *T. pretiosum*; 2) nine females of *T. pretiosum* and one female of *T. remus*; 3) eight females of *T. pretiosum* and two females of *T. remus*; 4) seven females of *T. pretiosum* and three females of *T. remus*; 5) six females of *T. pretiosum* and four females of *T. remus*; 6) five females of *T. pretiosum* and five females of *T. remus*; and 7) 10 females of *T. remus*.

After 24 h of parasitism, the cardboard pads with the parasitized eggs were removed from the vials and placed into transparent plastic bags, until the emergence of the adult parasitoids. The parameter evaluated was the percentage of parasitized eggs. Results were submitted to exploratory analysis to evaluate the assumptions of normality of residues, homogeneity of variance of treatments and additivity of the model to allow for ANOVA implementation. The means were then compared by the Tukey test at 5% probability.

RESULTS AND DISCUSSION

The treatments with 2 *T. remus* for each 8 *T. pretiosum* (20%) or higher proportion of *T. remus* presented the most effective parasitism of *S. frugiperda* eggs compared to the other treatments. The lowest percent parasitism was observed in the treatment where only *T. pretiosum* females were present (Fig. 1A). The greater efficiency in the treatments with *T. remus* is because this parasitoid is able to parasitize the eggs of the inner layers, affecting the entire egg mass (Fernandes & Carneiro 2006). On the other hand, this characteristic of *Spodoptera* spp. – oviposition in overlapping layers – provides a limitation to the parasitism by Trichogrammatidae. The impact of the number of *S. frugiperda* egg layers on the parasitism of *Trichogramma atopovirilia* Oatman & Platner has been previously reported in the literature, when the parasitism detected in single layer egg masses was higher than that observed in egg masses with two and three egg layers (Beserra & Parra 2005).

Differently, the usual limitation to the parasitism by Trichogrammatidae regarding to parasitism on *Spodoptera* spp. do not occur for *T. remus*. It is important to emphasize that the parasitism capacity of *T. remus* on eggs of *S. frugiperda* is considerably high (Bueno *et al.* 2008; Bueno *et al.* 2010). Each *T. remus* female produces about 270 offspring during their reproductive lifespan (Morales *et al.* 2000) and performs the oviposition of one egg per host, thus superparasitism is unusual in this species (Cave 2000). However, the superparasitism on eggs parasitized by *Trichogramma* spp. is not a rare event. In assessing parasitism and superparasitism of *T. pretiosum* on eggs of *Sitotroga cerealella* Olivier 1819 (Lepidoptera, Gelechiidae) a lower reproductive success of that parasitoid was observed (Moreira *et al.* 2009).

The same increment in efficiency observed for eggs of *S. frugiperda*, in relation to percentage of parasitism, was also observed for eggs of *S. cosmioides*, when *T. remus* was added to the treatments (Fig. 1B). That occurred because the arrangement of the *S. cosmioides* egg mass is similar to that of *S. frugiperda*, both being arranged in overlapping layers, hindering the action of *T. pretiosum*.

In contrast, for the parasitism of *S. eridania* eggs there was no statistical difference among the treatments with and without *T. remus*. It was found that even in the treatment with *T. pretiosum* alone, the parasitism was higher than 70% (Fig. 1C). It should be emphasized that the *S. eridania* eggs are not laid in overlapping layers, thus *T. pretiosum* can parasitize almost all eggs of the egg mass. This explains the good performance of parasitism with the release of only *T. pretiosum* females, which did not occur when the hosts were *S. frugiperda* and *S. cosmioides* eggs.

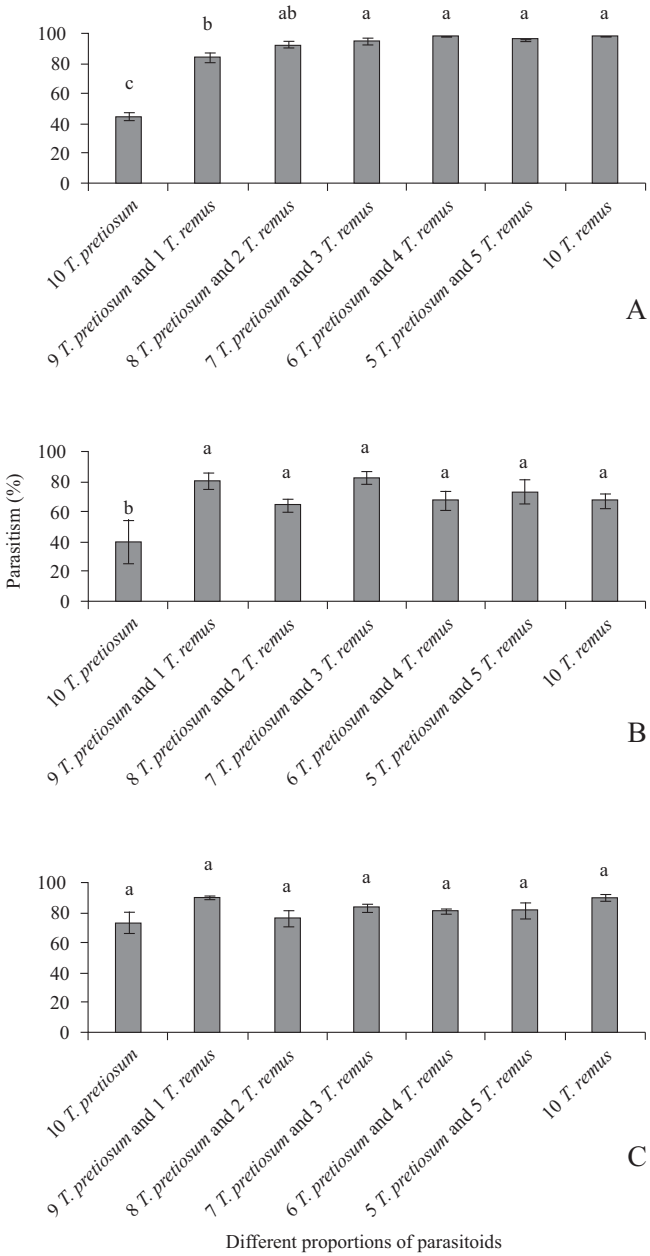

Fig. 1. Mean parasitism (± SEM) observed in different treatments using *Trichogramma pretiosum* and *Telenomus remus* in different proportions to parasitize egg masses of *Spodoptera frugiperda* (A), *Spodoptera cosmioides* (B) and *Spodoptera eridania* (C). Means followed by same letter are not statistically different by the Tukey test at 5% probability for each host evaluated.

The choice of the parasitoid to be released for controlling the *Spodoptera* complex on soybeans is very important, mainly because this complex of species has grown in importance in recent crop seasons (Bueno *et al.* 2009). Among the natural enemies of *Spodoptera* spp., the egg parasitoid *T. remus* stands out as the most common species of the genus, being already found parasitizing five different species of *Spodoptera* spp. (Morales *et al.* 2000). This parasitoid has been used in large-scale IPM programs in Venezuela, released in areas of corn, reaching rates of parasitism up to 90% on eggs of *S. frugiperda* (Ferrer 2001). If from one standpoint *T. remus* proved to be very efficient in the management of the *Spodoptera* complex, from another it is important to consider that on soybean crops, other caterpillars of economic importance also occur in association with *Spodoptera* spp, such as larvae of *P. includens* (Bueno *et al.* 2007). Therefore, the use of *T. remus* associated with *T. pretiosum* proves to be a good option in the biological control of soybean caterpillars. *T. pretiosum* was reported as the best species to be used against *P. includens* (Bueno *et al.* 2009). These larvae infest the crops during the reproductive stage and in addition to defoliation; they feed and damage the pods. During this period, the plant has a well-developed foliar architecture, thus impairing chemical control, which has often been ineffective due to the large amount of leaves that the plants have at that stage (Bueno *et al.* 2007).

Unlike the chemical control, the use of egg parasitoids can be more efficient because the parasitoid can search for the pest, even in a well developed soybean plant. In this case, the choice of the parasitoid species is of utmost importance. *T. pretiosum* has the advantage of being generalist, controlling several insect pests of the order Lepidoptera that attack soybean, such as *P. includens* (Bueno *et al.* 2009) and *A. gemmatalis* (Zachrisson & Parra 1998). Furthermore, the large-scale rearing of this parasitoid has lower production cost and is easier to carry out as compared to the multiplication of *T. remus*. However, *T. pretiosum* has the great disadvantage of not being efficient on pests that lay their eggs in overlapping, layers such as *S. cosmioides* and *S. frugiperda*. Despite being more efficient on these insect pests, *T. remus* is a parasitoid specific to the genus *Spodoptera* and would not have a good performance against other moths of economical importance on soybean crops, as *P. includens*, which occurs in the same period as *Spodoptera* spp. Thus, the combination of both is a good strategy for Lepidoptera control in soybean crops.

CONCLUSIONS

The parasitoid *T. pretiosum*, when released solely for parasitizing eggs of *S. frugiperda* and *S. cosmioides*, under controlled environmental conditions, is less efficient than when released associated with the parasitoid *T. remus*. Treatments combining both parasitoid species with only 1 *T. remus* for each 9 *T. pretiosum* (10%) and only 2 *T. remus* for each 8 *T. pretiosum* (20%) were sufficient to significantly increase

the parasitism up to rates statistically equivalent to the treatment with 100% of *T. remus* observed in eggs of *S. frugiperda* and *S. cosmioides*, respectively, under controlled laboratory conditions. For the biological control of *S. eridania*, the parasitoids *T. remus* and *T. pretiosum*, when released alone or in combination, under laboratory conditions, do not show statistically significant differences on percentage of parasitism. Experiments in the field should be carried out in the future to ratify these laboratory results for field conditions.

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