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cis-Jasmone indirect action on egg parasitoids (Hymenoptera: Scelionidae) and its application in biological control of soybean stink bugs (Hemiptera: Pentatomidae)

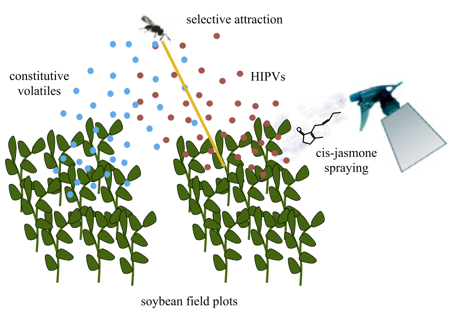
Cecília Rodrigues Vieira [a,b,*], Maria Carolina Blassioli Moraes [a], Miguel Borges [a], Edison Ryoiti Sujii [a,b], Raúl Alberto Laumann [a]

[a] *Embrapa Recursos Genéticos e Biotecnologia, Parque Estação Biológica, W5 Norte, Asa Norte, 70.770-917 Brasília, DF, Brazil*
[b] *Programa de Pós-Graduação em Ecologia, Universidade de Brasília, Campus Universitário Darcy Ribeiro, Asa Norte, 70.910-900 Brasília, DF, Brazil*

HIGHLIGHTS

- *cis*-Jasmone is an herbivore induced plant volatile.
- *cis*-Jasmone may induce indirect defenses in soybean plants against stink bugs.
- Sprayed *cis*-jasmone increased number of Scelionidae egg parasitoids in soybean plots, when the matrix is soybean.
- Sprayed *cis*-jasmone did not change parasitism and number of stink bugs in soybean plots.

GRAPHICAL ABSTRACT



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ABSTRACT

In many plants, the secondary metabolite *cis*-jasmone activates the metabolic pathway that produces volatile organic compounds attractive to natural enemies and, sometimes, repellent to herbivores. Previous studies indicate that the feeding damage caused by the herbivore *Euschistus heros* or the exogenous application of *cis*-jasmone in soybean plants induces the release of herbivore-induced plant volatiles (HIPVs) with a similar chemical profile and these compounds can attract the stink bug parasitoid *Telenomus podisi* (Scelionidae). Herein we tested in field conditions the effect of exogenous application of *cis*-jasmone in soybean plants on the parasitoid and stink bug community and on stink bug egg parasitism. In two areas, one within a soybean and another within a *Crotalaria* matrix, we randomly distributed 2 m^2 plots, with soybean plants induced (treatment, $n = 5$) or not induced by *cis*-jasmone (control, $n = 5$) in the field. We sampled the parasitoid community weekly with yellow sticky traps ($n = 3$/plot) and monitored parasitism with sentinel eggs of *E. heros* ($n = 150$/plot). We also monitored the population of stink bugs weekly, by sampling each plot with shake-cloth technique. The abundance of Scelionidae was highest overall and also in treated plots during the first four weeks in the area with a soybean matrix, but decreased thereafter. The richness of parasitoid families was similar between treatment and control plots in the area with a soybean matrix, but higher in control plots in the area with a *Crotalaria* matrix. Evenness was higher in control plots in the area with soybean matrix, whereas the reverse occurred in the area with a *Crotalaria* matrix. Results suggest that treatment with *cis*-jasmone effectively attracted and enhanced the population of scelionid parasitoids, but had no effect on the occurrence and intensity of parasitism and in the number of stink bugs.

* Corresponding author. Present address: Departamento de Zoologia, Universidade de Brasília, Campus Universitário Darcy Ribeiro, Asa Norte, 70.910-900 Brasília, DF, Brazil. Fax: +55 61 3202 2039.

E-mail addresses: ceciliavieira@me.com (C.R. Vieira), carolina.blassioli@embrapa.br (M.C.B. Moraes), miguel.borges@embrapa.br (M. Borges), edison.sujii@embrapa.br (E.R. Sujii), raul.laumann@embrapa.br (R.A. Laumann).

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1. Introduction

Over evolutionary time, plants developed many mechanisms to resist adverse conditions, including the production of secondary metabolites against herbivores and pathogens (Baldwin, 2010; Dicke et al., 2003). Herbivore-induced plant volatiles (HIPVs) are synthesized in plant tissues and released in response to herbivory (Dudareva et al., 2006; Hilker and Meiners, 2006). Besides repelling herbivore insects, HIPVs indirectly attract herbivore's natural enemies, providing information on the location of hosts and prey and protecting plants from subsequent attacks (Dudareva et al., 2006; Vet and Dicke, 1992).

HIPVs are complex blends of compounds, including green leaf volatiles (GLVs), aromatic compounds, mono-, homo- and sesquiterpenes. After herbivore attack, the production of HIPVs depends on metabolic pathways activated by different compounds, such as phytohormones or their derivatives (Baldwin, 2010; Dudareva et al., 2006; van Den Boom et al., 2004). When applied on plants, the naturally obtained compounds or their synthetic standards act as elicitors of HIPVs (Heil and Walters, 2009). For instance, the application of jasmonic acid on different plants promotes the emission of volatiles attractive to parasitoids (de Moraes et al., 2000; Heil, 2004; Pickett et al., 2007). Likewise, the application of *cis*-jasmone, a metabolite derived from the biosynthesis of jasmonic acid (octanodecanoid pathway, Birkett et al., 2000), induces defenses in wheat that repel pest aphids and increase their attractiveness to parasitoids (Bruce et al., 2003b). In soybean plants, *cis*-jasmone promotes the liberation of HIPVs similar to those induced by stink bug (Pentatomidae) injuries (Moraes et al., 2005, 2008), and these HIPVs attract the egg parasitoid *Telenomus podisi* (Ashmead) (Moraes et al., 2009). Thus, the induction of HIPV emission can be used in the behavioral manipulation of parasitoids that naturally occur in and around crops, or parasitoids released for augmentative biological control (Powell and Pickett, 2003). Nevertheless, an understanding of the impact of HIPV elicitors on the community structure of insect parasitoids is still wanting. This knowledge is essential to establish a solid scientific basis for the management of insect parasitoids in crops.

In Brazil, stink bugs are one of the most serious pests of soybean plantations (Panizzi, 1997). The biological control of these herbivores by egg parasitoids is an alternative that could reduce the use of synthetic insecticides and contribute to the conservation of natural resources, becoming an important tool for sustainable agriculture (Corrêa-Ferreira and Moscardi, 1996). Herein we evaluate in the field, the effects of induced defenses in soybean plants treated with *cis*-jasmone on the attraction of Scelionidae parasitoids of stink bug eggs in particular, and on the abundance and richness patterns of parasitoid community in general. We expected an increase in the density and richness of parasitoids in treated plots, especially scelionids, and higher prevalence and intensity of parasitism in stink bug eggs.

2. Material and methods

2.1. Study site

We conducted field experiments in two adjacent areas at Embrapa Recursos Genéticos e Biotecnologia, Distrito Federal, Brazil (15°43′50″ S, 47°53′59″ W). One consisted of 0.55 ha of soybean, *Glycine max* (L.) Merr. (cv-MSOY 6101; hereafter called "soybean area"), whereas the other had a total area of 0.71 ha of rattle pod, *Crotalaria juncea* L. (hereafter called "*Crotalaria* area"). Within each area, we randomly embedded 10 small plots with soybean (cv BR-16) between November 2008 and March 2009. We fertilized the soil, corrected its superficial acidity and inoculated seeds with nitrogen fixing bacteria (*Bradyrhizobium* sp.) at a rate of 1 kg of inoculant to 50 kg of seeds following recommendations of Embrapa (2010).

2.2. Experimental design

In each area, we conducted experiments in the early reproductive stage of soybean plants in 10 randomly distributed plots. Each plot measured 2.0×1.0 m and they were far between at least 10 m. We manually sprayed soybean plants of five plots (treatment) in each area with *cis*-jasmone (TCI, Japan), using 6.0 ml/plant of a solution of 250 mg of *cis*-jasmone and 100 mg of surfactant Tween® 20 (Sigma–Aldrich, United States) diluted in 1 l of water. We based the dosage of solution and procedures of application on a previous laboratory study carried with *cis*-jasmone and the same tritrophic interaction of this study (Moraes et al., 2009). We sprayed soybean plants in the initial reproductive stage (R1-flowering, Ritchie et al., 1988), when the crop is colonized by stink bugs (Panizzi, 1997), and maintained plots until the physiological maturity of soybean, when stink bugs migrate to refuges or areas with alternative host plants.

2.3. Sampling of parasitoids and stink bugs

We sampled parasitoids during nine weeks with 15.0×12.5 cm rectangles of yellow sticky traps (Biotrap®, BioControle Métodos de Controle de Pragas, Brazil), attached by wire to wooden stakes about 1 m high from the ground. In each plot, we placed traps in three points along one of the diagonals, one point at the center and one at each edge of the diagonal. The first sampling was synchronized with the spraying of plants. The adhesive surface of traps was exposed in the field for three days, after which they were removed, wrapped in plastic film, and stored in refrigerator for the identification of insects. We replaced traps every week. We identified Scelionidae parasitoids to the lowest possible taxonomic rank, following the dichotomous key developed by Dr. Marta Loiácono (División Entomología del Museo de La Plata, Argentina), and other parasitoids to the family level following Gauld and Bolton (1988) and Goulet and Huber (1993).

We recorded the incidence (presence or absence) and intensity (number of parasitized eggs/number of remaining eggs) of egg parasitism weekly, by randomly placing three cardboard squares in each plot (3.0×3.0 cm), each cardboard square containing 50 sentinel eggs held by Arabic gum. We covered each cardboard square with tulle (silk net) ~2.0 mm diameter to avoid predation; the tulle allowed free passage of parasitoids but precluded access to eggs by predators (e.g., bugs, ants, and beetles). We obtained eggs of the brown stink bug, *Euschistus heros* (Fabricius), the main soybean pest in central Brazil, from a colony maintained at Embrapa Recursos Genéticos e Biotecnologia, following the rearing procedures described in Laumann et al. (2011). In each plot, we placed cardboards with eggs on three different plants, tied with color tape on a leaf petiole. Eggs were exposed in the field for three days and then transported to the laboratory, packed in plastic pots with lids, and maintained in a room with controlled environmental conditions (LD 14:10 h, 26.0 ± 1 °C and 65 ± 10% relative humidity). We verified parasitism by the emergence of parasitoids or by dissecting eggs and recording the presence of pupae and immature stages, when parasitoids did not complete their development. We counted parasitized eggs and identified emerged parasitoids under a Zeiss SV6 stereomicroscope. Of the 270 sentinel egg cards placed in each study area, 107 (soybean: 40%) and 152 (*Crotalaria*: 56%) were lost by the action of rain, predation (e.g., ants, beetles and other stink bugs) or mastication of the card with the tulle (e.g., Orthoptera), and these cards were not considered in statistical analyses.