Integrated control of two tortricid (Lepidoptera) pests in apple orchards with sex pheromones and insecticides

Control integrado de dos tortricidos (Lepidoptera) plaga en huertos de manzanos con feromonas sexuales e insecticidas

PATRIK LUIZ PASTORI[1], CRISTIANO JOÃO ARIOLI[2], MARCOS BOTTON[3], LINO BITTENCOURT MONTEIRO[4], LYNDSIE STOLTMAN[5] and AGENOR MAFRA-NETO[6]

Abstract: The apple is attacked by a significant number of insect pests in Brazilian commercial orchards, including *Bonagota salubricola* and *Grapholita molesta* (Lepidoptera: Tortricidae). Sexual disruption of *B. salubricola* and *G. molesta* was evaluated in apple orchard using the flowable pheromone formulations, SPLAT Grafo+Bona (SG+B), SPLAT Attract and Kill Grafo+Bona (SAKG+B), and compared with the standard insecticides used for management in the Integrated Apple Production (IAP) system. Both formulations were applied at a rate of 1kg/ha on October 10, 2005 and December 13, 2005 using 300 and 1000 point sources/ha of SG+B and SAKG+B, respectively in experimental units of 7 ha. Adult male captures of *B. salubricola* and *G. molesta* were evaluated weekly in Delta traps with specific synthetic sex pheromone from October 10, 2005 to February 14, 2006. Damage to fruits was evaluated on November 21 and December 21, 2005, and January 25 and February 14, 2006. In the SPLAT treated experimental units a significant reduction was observed in the number of *B. salubricola* and *G. molesta* males caught in Delta traps compared to the experimental unit IAP. Damage by *B. salubricola* at harvest ranged from 1.63 to 4.75% with no differences between treatments, while damage by *G. molesta* was near zero in all experimental units. Mating disruption using SG+B and SAKG+B was sufficient to control *B. salubricola* and *G. molesta* with results equivalent to IAP guidelines. This technology is promising for management of both pests in Brazilian apple orchards with immediate reduction of 43% in the number of insecticide applications.

Key words: Attract and kill. Integrated Apple Production. Lepidoptera. Mating disruption. SPLAT.

Resumen: Las plantaciones comerciales brasileñas de manzana son atacadas por un gran número de insectos plaga, entre ellas *Bonagota salubricola* y *Grapholita molesta* (Lepidoptera: Tortricidae). La interrupción del apareamiento de *B. salubricola* y *G. molesta* fue evaluada en huerto de manzanas usando las formulaciones de feromonas pastosa, SPLAT Grafo + Bona (SG+B) y SPLAT Attract y Kill Grafo + Bona (SAKG+B), y comparado con los insecticidas estándares que se usan para el manejo en la Producción Integrada de Manzanas (PIM). Ambas formulaciones se aplicaron a una tasa de 1kg/ha el 10 de octubre de 2005 y el 13 de diciembre 2005 usando 300 y 1.000 fuentes puntuales /ha de SG+B y SAKG+B, respectivamente, en unidades experimentales de 7 ha. La captura de adultos machos de *B. salubricola* y *G. molesta* fue evaluada semanalmente en trampas Delta con feromona sexual sintética específica del 10 de octubre de 2005 al 14 de febrero de 2006. Los daños en las frutas se evaluaron el 21 de noviembre y 21 de diciembre de 2005, y el 25 de enero y 14 de febrero de 2006. En las unidades experimentales tratadas con SPLAT se observó una reducción significativa en el número de machos de *B. salubricola* y *G. molesta* capturados en trampas Delta en comparación con la unidad experimental PIM. El daño causado por *B. salubricola* en la cosecha varió desde 1,63 hasta 4,75%, sin diferencias entre los tratamientos, mientras que los daños causados por *G. molesta* estaban cerca de cero en todas las unidades experimentales. La interrupción del apareamiento con SG+B y SAKG+B fue suficiente para controlar *B. salubricola* y *G. molesta* con resultados equivalentes a las directrices de la PIM. Esta tecnología es promisoria para el manejo de ambas plagas en huertos de manzanas brasileños con reducción inmediata del 43% en el número de aplicaciones de insecticidas.

Palabras clave: Atraer y matar. Producción integrada de manzanas. Lepidoptera. Interrupción del apareamiento. SPLAT.

Introduction

The apple, *Malus domestica* Borkhausen, 1760 (Rosaceae), has a number of phytosanitary problems, with emphasis on the occurrence of insect pests that significantly reduce yield. The Brazilian apple leafroller, *Bonagota salubricola* (Meyrick, 1937) (Lepidoptera: Tortricidae) and the Oriental fruit moth, *Grapholita molesta* (Busck, 1916) (Lepidoptera: Tortricidae), stand out among the main apple pests (Arioli *et al.* 2007; Botton *et al.* 2009) causing significant losses if control measures are not adopted (Botton *et al.* 2000a). *B. salubricola* larvae feed on apple leaves and fruits, causing severe damage and economic losses (Botton *et al.* 2000a, 2009). Damage from *G. molesta* moths can be seen on shoots and fruits, causing significant losses when the attack occurs in nurseries, orchards, and cultivars with greater fruit production in terminal buds, such as 'Fuji' (Arioli *et al.* 2007).

Bonagota salubricola and *G. molesta* have been primarily managed with insecticides (Arioli *et al.* 2004; Botton *et al.* 2000a, 2009), which can leave harmful residues on fruits (Thomson *et al.* 2001). New control alternatives need to be studied, especially when the crop is managed by Integrated Apple Production (IAP) system, this is a new approach production which targets consumer concerns over the impact

[1] Ph. D. Departamento de Fitotecnia, Universidade Federal do Ceará, 60.356-000, Fortaleza, Ceará, Brazil. *plpastori@ufc.br,* corresponding author. [2] Ph. D. Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina, Estação Experimental de Videira, 89.560-000, Videira, Santa Catarina, Brazil, *cristianoarioli@epagri.sc.gov.br.* [3] Ph. D. Centro Nacional de Pesquisa Uva e Vinho, Empresa Brasileira de Pesquisa Agropecuária, 95.700-000, Bento Gonçalves, Rio Grande do Sul, Brazil, *marcos@cnpuv.embrapa.br.* [4] Ph. D. Departamento de Fitotecnia e Fitossanitarismo, Universidade Federal do Paraná, 80.035-050, Curitiba, Paraná, Brazil, *lbmonteiro@terra.com.br.* [5] B.S. ISCA Technologies Inc., 1230 Spring Street, Riverside CA 92507, USA. *lyndsie.stoltman@iscatech.com.* [6] Ph. D. ISCA Technologies Inc., 1230 Spring Street, Riverside CA 92507, USA. *president@iscatech.com.*

of crop production practices on the environment. The pest management philosophy embodied in Integrated Fruit Production (IFP) requires greater emphasis on the use of biological control, pest thresholds, minimal use of broad-spectrum pesticides and replacement with selective products (Protas and Sanhueza 2002). One alternative for the management of insect pests is the use of sex pheromones: chemicals that are used for mating communication between both sexes of the same species (Karlson and Luscher 1959) that can be produced by males or females (Cardé and Minks 1995). These substances can be synthesized and used for insect monitoring or control by mating disruption or attract and kill strategies (Cardé and Minks 1995; Bosa *et al.* 2008; Pastori *et al.* 2008).

The availability of new pheromone formulations to control *G. molesta* allows for a wide use of the technology in *M. domestica* (Monteiro *et al.* 2008; Pastori *et al.* 2008). However, where this technology is employed, there is a need for insecticide applications against other pests, particularly the South American fruit fly, *Anastrepha fraterculus* (Wiedemann, 1830) (Diptera: Tephritidae) (Monteiro *et al.* 2008). The sex pheromone of *B. salubricola* has been isolated, identified (Unelius *et al.* 1996; Coracini *et al.* 2001), and is commercially used for monitoring purposes (Kovaleski *et al.* 2003; Botton *et al.* 2009). Specialized Pheromone & Lure Application Technology, or SPLAT®, allows for the combination of multiple pheromones into a single formulation, as for *B. salubricola* and *G. molesta,* expanding the possibility for simultaneous control of these Lepidopteran pests.

The global market offers a variety of pheromone products for control of mainly Lepidopteran pests. The various products available differ in their formulations and in their costs and times of application (Degen *et al.* 2005; Stelinski *et al.* 2005; Epstein *et al.* 2006; Miller *et al.* 2006a, b). Most products are applied by hand, requiring an intensive labor force (Jenkins 2002; Stelinski *et al.* 2005, 2006). Microencapsulation of pheromone formulations is one alternative to minimize labor, using sprinklers or irrigation systems for application. However, several reapplications are required over the crop cycle because of the short residual effect (Knight *et al.* 2004; Botton *et al.* 2005) and resistance problems (Suckling *et al.* 1999). Because SPLAT is an inert wax matrix, it allows the user the flexibility to control the pheromone release in the field by varying the number and size of point sources (Miller *et al.* 2006a, b; Mafra-Neto *et al.* 2008). In addition, the formulation is not washed away by rain and can be adapted to many different forms of application, from small-scale hand application (calibrated dosing guns, caulking tubes, pastry bags, etc.) to mechanical application for large areas (tractor equipment), which maximizes efficiency by decreasing the application cost (Stelinski *et al.* 2005, 2006; Mafra-Neto *et al.* 2008).

Sex pheromones are species-specific (Degen *et al.* 2005); the presence of other species that are not controlled with pheromones can still result in severe damage, hampering the use of this technology. One alternative is the application of sex pheromones to control key pests along with a reduced number of chemical insecticide applications for other species (Meissner *et al.* 2001; Trimble *et al.* 2001; Kovanci *et al.* 2005). The objectives of this study were to evaluate the integrated control of *B. salubricola* and *G. molesta* based on 1) the timing, quantity, and spacing of SPLAT treatments and 2) the mechanism of semiochemical control (i.e. mating disruption versus attract and kill).

Materials and Methods

The experiment was carried out from October 2005 to February 2006 in an apple orchard in the municipality of Vacaria, Rio Grande do Sul State, Brazil.

Experimental site and treatments. The experiment was set in apple orchards established in 2001 with a spacing of 1.5 x 4.5 m (plants x rows) and tree height between 2.5 and 3.0 m. The orchards contained a combination of four rows of 'Gala' (producer) for every two rows of 'Fuji' (pollenizer) varieties. The orchards were divided into five experimental units of seven hectares each, spaced at least 200 m apart in order to prevent migration of mated females. Each experimental unit received one of the following treatments: A) SPLAT Grafo + Bona (SG+B) at a rate of 1 kg/ha (300 point sources, each 3.3 g) applied on 10/04/05, B) SPLAT Grafo + Bona (SG+B) at a rate of 1 kg/ha (300 point sources, each 3.3 g) applied on 12/13/05, C) SPLAT Attract and Kill Grafo + Bona (SAKG+B) at a rate of 1 kg/ha (1000 point sources, each 1 g) applied on 10/04/05, D) SPLAT Attract and Kill Grafo + Bona (SAKG+B) at a rate of 1 kg/ha (1000 point sources, each 1 g) applied on 12/13/05, and E) pest management under recommendation of the Integrated Apple Production (IAP) (Protas and Sanhueza 2002) with the following treatments: tebufenozide (Mimic SC 240, 90 mL/100L) on 10/25/05 for *B. salubricola* and *G. molesta*, phosmet (Imidan 500 PM, 120 g/100L) on 11/07/05 for *A. fraterculus*, fenitrothion (Sumithion 500, 150 mL/100L) on 11/18/05 and 12/05/05 for *A. fraterculus* and *G. molesta*, methidation (Supracid 400, 100 mL/100L) on 12/20/05 and 01/08/06 for *A. fraterculus*, and chlorpyrifos (Lorsban 480 CE 120 mL/100L) on 01/03/06 for *B. salubricola* and *G. molesta*. Three insecticide applications were carried out in each experimental unit where SPLAT was applied, on the basis of adult monitoring for the control of *A. fraterculus* as follows: phosmet (Imidan 500, 120 g/100L) on 11/07/05 and methidathion (Supracid 400, 100 mL/100L) on 12/20/05 and 01/12/06. The risk of damage to production and the size of areas needed to set up the experiment did not allow for an experimental unit without insecticide application and replication the experimental units of the seven hectares each.

Synthetic sex pheromone formulation. SPLAT, developed and patented by ISCA Technologies (Riverside, California USA), is a flowable, amorphous emulsion consisting of oils and waxes that controls semiochemical release. SPLAT Grafo + Bona and SPLAT Attract and Kill Grafo + Bona are made up of a mixture of sex pheromones from *B. salubricola* and *G. molesta* including: **SG+B** - [(E)-8-dodecenyl acetate; (Z)-8-dodecenyl acetate; (Z)-8-dodecenol (4.4%) (44 g/kg); (E, Z)-3.5-dodecadienyl acetate (0.20%) (2 g/kg)], and **SAKG+B** - [(E)-8-dodecenyl acetate; (Z)-8-dodecenyl acetate; Z-8-Dodecenol (2.2%) (22 g/kg); (E, Z)-3,5-dodecadienyl acetate (0.20%) (2 g/kg); (RS)-alpha-cyano-3-phenoxybenzyl (1RS,3RS;1RS,3SR)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate (cypermethrin) (5%) (50 g/kg)].

SPLAT application. SPLAT was applied by hand in each experimental unit using wooden spatulas adjusted to hold

3.3 g (SG+B) and 1 g (SAKG+B), applying the formulation at a dose of 1 kg/ha. Ten percent more SPLAT was applied at the inner borders of each treatment (approximately 10 m) to reduce edge effects (Albajes *et al*. 2002; Degen *et al*. 2005). SPLAT was applied on shaded portions of plants, at the base of the branches between 1.5 and 2.0 m above ground.

Preliminary evaluation of pest population. Five white Delta traps baited with Iscalure *Bonagota*® and five others with Iscalure *Grapholita*® (ISCA Tecnologias Ltda., Ijuí, RS, Brazil) were placed in each of the experimental units on plants 30 m apart, between 1.5 and 2.0 m above ground, on 09/20/05. These traps were evaluated seven days later to determine the homogeneity of experimental units and initial population levels of *B. salubricola* and *G. molesta*.

Treatment evaluation. Treatment efficacy was assessed by recording the adult population and damage to fruit by *B. salubricola* and *G. molesta*. Male moth captures in Delta traps were evaluated weekly from the beginning of the experiment. *B. salubricola* and *G. molesta* lures in Delta traps were replaced every 90 and 30 days respectively, and the adhesive trap bottoms were replaced as needed.

Mating disruption was evaluated by calculating the mating interruption rate (MIR), with the formula MIR= (C-T/T)*100, where "C" is the average of males captured per trap in the experimental unit under treatment and "T" is the number of catches in the Integrated Apple Production (IAP) (Molinari *et al.* 2000). MIR was calculated using the entire period of the experiment.

Damage by pests in apple fruits was evaluated on four dates (11/21/05, 12/21/05, 1/25/06 and 2/14/06) by recording the number of fruits damaged by *B. salubricola* and *G. molesta* from a sample of 1600 fruits per treatment (eight sampling locations with 200 fruits per location).

Data analysis. The population fluctuation of adult male of *B. salubricola* and *G. molesta* by plotting the average number of males/trap/week as a function of the time per each treatment.

The mean number of moths for each experimental unit was compared from 10/04/05 to 02/14/06. The percentage of damaged fruit was compared by transforming data by $\sqrt{(x+0.5)}$. Data was examined by analysis of variance and means were compared using Tukey's test, with a significance level of 0.05.

Results

The mean number of adults of *B. salubricola* (F = 1.2368; df = 20.0; P = 0.3271) and *G. molesta* (F = 2.3333; df = 20.0; P = 0.0907) captured in Delta traps in the preliminary evaluation (09/27/05) was statistically similar among the experimental units, demonstrating homogeneity of infestation (Table 1).

The mean number of male *B. salubricola* captured in the experimental units treated with SPLAT on either 10/04/05 or 12/13/05 was significantly lower than that observed in the Integrated Apple Production (IAP) treated experimental unit (F = 14.5002; df = 20.0; P = 0.0001) (Table 1). In the experimental units treated on 10/04/05, the mating interruption rate (MIR) of *B. salubricola* was 84.4 and 75.5% for SG+B and SAKG+B respectively. SPLAT applied on 12/13/05 resulted in lesser MIRs; 66.1 and 65% for SG+B and SAKG+B, respectively (Table 1). These results indicate that the SPLAT application on 10/04/05 was more effective for mating disruption than the application on 12/13/05.

MIR's for *G. molesta* were above 90% when SG+B and SAKG+B were applied on 10/04/05 (Table 1) was significantly lower than that observed in the other treatments (F = 33.5565; df = 20.0; P = 0.0001). Applying SPLAT on 12/13/05 reduced the MIR's to just 52.1 and 75.1% for SG+B and SAKG+B, respectively (Table 1).

The fluctuation of male populations of *B. salubricola* and *G. molesta* after SPLAT application on 10/04/05 was significantly lower than that observed in the Integrated Apple Production (IAP) system treated areas during the experiment (Fig. 1). The fluctuation of male *B. salubricola* and *G. molesta* populations in the experimental units treated on 12/13/05 showed similar behavior to the Integrated Apple Production (IAP) treated experimental unit before the SPLAT treatments were applied, and was significantly reduced after the SPLAT applications (Fig. 1).

Damage to fruits caused by *B. salubricola* and *G. molesta* on 11/21/05 (first evaluation) (*B. salubricola* F = 1.000; df =

Table 1. Males (μ ± SE) of *Bonagota salubricola* and *Grapholita molesta* collected and mating interruption rate (MIR%) in plots of apple trees treated with: SPLAT Grafo + Bona (SG+B) and SPLAT Attract and Kill Grafo + Bona (SAKG+B) applied on 10/04/05 or 12/13/05, and Integrated Apple Production (IAP) system. Vacaria, Rio Grande do Sul State, Brazil, 2005/2006.

Treatment	*Bonagota salubricola*			*Grapholita molesta*		
	Pre-Evaluation[1]	μ Total[2]	MIR (%)[3]	Pre- Evaluation[1]	μ Total[2]	MIR (%)[3]
SG+B (10/04/05)	1.0 ± 0.28	28.2 ± 5.58 a	84.4	0.0 ± 0.00	1.2 ± 0.37 a	98.7
SG+B (12/13/05)	1.0 ± 0.28	61.2 ± 11.37 a	66.1	0.4 ± 0.22	45.4 ± 7.33 b	52.1
SAKG+B (10/04/05)	1.6 ± 0.46	44.2 ± 7.93 a	75.5	0.2 ± 0.18	7.6 ± 3.08 a	92.0
SAKG+B (12/13/05)	0.4 ± 0.22	63.2 ± 9.26 a	65.0	0.0 ± 0.00	23.6 ± 6.11 ab	75.1
IAP	1.2 ± 0.46	180.6 ± 30.84 b	–	0.8 ± 0.33	94.8 ± 10.58 c	–

[1] Non-significant by F test (P = 0.05).
[2] Means followed by the same small letter in the column are not statistically different by the Tukey test (P = 0.05).
[3] MIR = (Pheromone - IAP / IAP)*100.

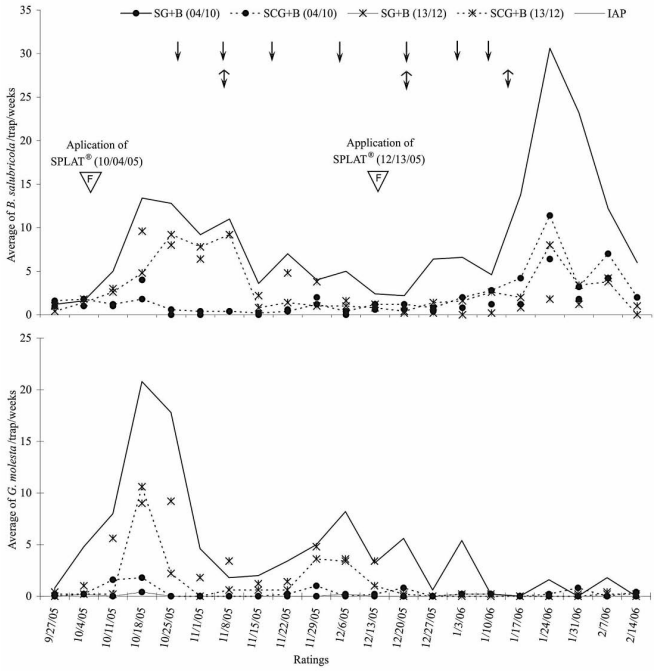

Figure 1. Average number of male *B. salubricola* and *G. molesta* collected per week in Delta traps baited with Iscalure *Bonagota* and Iscalure *Grapholita* in plots of apple trees treated with: SPLAT Grafo + Bona (SG+B) and SPLAT Attract and Kill Grafo + Bona (SAKG+B) applied on 10/04/05 or 12/13/05 in different experimental units, and Integrated Apple Production (IAP) system. Seven arrows standard indicate application of insecticides in experimental unit Integrated Apple Production (IAP) and three arrows differentiated indicate application of insecticides in experimental units treated witch pheromones. Vacaria, RS, Brazil, 2005-2006.

35.0; P = 0.4206 and *G. molesta* F = 1.0000; df = 35.0; P = 0.4206 and subsequent evaluations on 12/21/05 (*B. salubricola* F = 1.5858; df = 35.0; P = 0.1998 and *G. molesta* F = 1.5757; df = 35.0; P = 0.2024) and 01/25/06 (*B. salubricola* F = 0.2549; df = 35.0; P = 0.9045 and *G. molesta* F = 1.5757; df = 35.0; P = 0.2024), in the experimental units treated with SPLAT, was statistically similar to that observed in the Integrated Apple Production (IAP) system treated experimental unit, ranging from 0 to 1.38% of apples damaged by *B. salubricola* and 0 to 0.38% for *G. molesta* (Table 2). For the final evaluation performed on 02/14/06, the fruit damage ranged from 1.63 to 4.75% and 0 to 0.13% for *B. salubricola* (F = 2.4849; df = 35.0; P = 0.0613) and *G. molesta* (F = 0.7500; df = 35.0; P = 0.5647) respectively, showing no significant differences (Table 2).

Discussion

Reduction in catches of male *B. salubricola* and *G. molesta* in the SPLAT treated experimental units compared to the Integrated Apple Production (IAP) system was the result of SPLAT formulations that disoriented males and prevented them from locating the Delta traps; a result reportedly observed when the same SPLAT technology was used for the control of *G. molesta* (Stelinski *et al.* 2005; Monteiro *et al.* 2008), *Paralobesia viteanea* (Clemens, 1860) (Lepidoptera: Tortricidae) (Jenkins and Isaacs 2008) and *Tecia solanivora* (Povolny, 1973) (Lepidoptera: Gelechiidae) (Bosa *et al.* 2008).

The late application of SPLAT was not the best strategy for increasing MIRs, given that in the second application these rates were lower than in the first. Apple producers have accepted yield losses by pests of 1 to 2% at harvest, which would represent about 400 to 800 kg of apples given a production of 40 t/ha. In this case, the application of pheromone in October was more promising from the practical viewpoint for managing *B. salubricola* and *G. molesta*.

We expected to see a direct relationship between reduction in the number of males captured in Delta traps and fruit damage, but this was not the case in the experimental units treated with SPLAT or the Integrated Apple Production (IAP) system. The experimental unit receiving the Integrat-

Table 2. Apples (% ± SE) damaged by *Bonagota salubricola* and *Grapholita molesta* in four evaluations in experimental units treated with: SPLAT Grafo + Bona (SG+B) and SPLAT Attract and Kill Grafo + Bona (SAKG+B) applied on 10/04/05 and 12/13/05, and Integrated Apple Production (IAP) system. Vacaria, Rio Grande do Sul State, Brazil, 2005/2006.

Treatments	Evaluations			
	11/21/05	12/21/05	01/25/06	02/14/06
	Percentage of damaged apples (*Bonagota salubricola*)[ns]			
SG+B (10/4/05)	0.00 ± 0.00	0.50 ± 0.38	0.75 ± 0.62	1.63 ± 0.86
SG+B (12/13/05)	0.00 ± 0.00	0.00 ± 0.00	1.38 ± 0.60	4.75 ± 0.75
SAKG+B (10/4/05)	0.00 ± 0.00	0.00 ± 0.00	1.00 ± 0.63	2.63 ± 0.96
SAKG+B (12/13/05)	0.00 ± 0.00	0.25 ± 0.25	0.88 ± 0.44	3.63 ± 1.03
IAP	0.00 ± 0.00	1.00 ± 0.63	1.25 ± 0.62	2.25 ± 0.70
	Percentage of damaged apples (*Grapholita molesta*)[ns]			
SG+B (10/4/05)	0.00 ± 0.00	0.38 ± 0.26	0.38 ± 0.26	0.13 ± 0.13
SG+B (12/13/05)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
SAKG+B (10/4/05)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
SAKG+B (12/13/05)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
IAP	0.13 ± 0.13	0.13 ± 0.13	0.13 ± 0.13	0.13 ± 0.13

[ns] Non-significant by F test (P = 0.05).

ed Apple Production (IAP) system treatment saw the greatest number of males caught in traps during the experiment, with damage similar to the other treatments at harvest. Traps baited with sex pheromone provided limited information about the movement patterns and density of males in the agro-ecosystem and did not consider females and sex ratio in the sampled population (Charmillot and Vickers 1991). Consequently, trap catch data may not always coincide with damage levels.

The lower efficiency of pheromone applied in December was caused by factors related to behavior and population dynamics of the target species (Cardé and Minks 1995; Moschos *et al.* 2004), which are still unclear for *B. salubricola*, an insect pest native to South America (Pastori *et al.* 2007). *B. salubricola* has no diapause and shows overlapping of generations (Botton *et al.* 2000b), that difficult to define the moment of pheromone treatment in the field. The delay in SPLAT applications increased the probability of random mating in the orchard. The large number of males in the treated areas facilitated accidental encounters between males and females, and increased the mating percentage which, consequently, reduced the SPLAT efficiency (Michereff Filho *et al.* 2000). The polyphagous habit of *B. salubricola* (Botton *et al.* 2009) allows fertilized females to migrate from nearby infested areas, while *G. molesta* is limited by a lower range of alternative hosts. Flight activity between apple orchards and adjacent ecosystems was important (Jeanneret and Charmillot 1995) and this accounts for the increase in damage caused by *B. salubricola* compared to *G. molesta*. The main factors related to the efficiency of semiochemicals for the control of *G. molesta* have not yet been studied in Brazilian fruit production (Molinari 2002), especially in relation to male population density and minimum area required to avoid edge effects (Albajes *et al.* 2002; Degen *et al.* 2005).

Reductions in the fluctuation of populations of male *G. molesta* in the areas treated with SPLAT over the course of this study and the percentage of damage to fruit, were generally similar to those reported in other countries in apple and peach crops (Vickers *et al.* 1985; Ricciolini and Baldi 1990; Molinari *et al.* 2000, 2004; Cravedi *et al.* 2001; Angeli *et al.* 2003; Il'ichev *et al.* 2004). Moreover, mating disruption with SPLAT has been successfully used to control *G. molesta* in apple (Stelinski *et al.* 2005, 2006, 2007) and *P. viteanea* in grapevines in the U.S. (Jenkins and Isaacs 2008).

Promising results with the use of sex pheromones in Brazil were reported for *G. molesta* in peach (Salles and Marini 1989; Botton *et al.* 2005) and apple (Monteiro *et al.* 2008; Pastori *et al.* 2008). In these cases, the specificity of the compounds has restricted the use of the technology, showing that the strategy of combining pheromones of more than one species in the same formulation and/or the combined use of insecticide appears to be promising for the widespread adoption of sex pheromones for mating disruption (Meissner *et al.* 2001; Trimble *et al.* 2001; Kovanci *et al.* 2005, Simon *et al.* 2007), allowing a reduction in insecticide use (Ricciolini and Baldi 1990; Atanassov *et al.* 2003) with less environmental degradation.

The SPLAT pheromone treatment allows growers to harvest product with no residue, allowing them to sell the product in the global market (especially being able to reach the European and Japanese markets).

Conclusions

The use of SPLAT Grafo + Bona or SPLAT Attract and Kill Grafo - Bona, together with three applications of insecticides to control *A. fraterculus,* proved to be an effective strategy for the management of *B. salubricola* and *G. molesta* in apple orchards, producing results similar to those of the Integrated Apple Production system, but with the use of far less insecticide.

SPLAT Grafo + Bona and SPLAT Attract and Kill Grafo + Bona are specific to *B. salubricola* and *G. molesta*, but the presence of other phytophagous species damaging the apple tree, particularly *A. fraterculus,* larvae of Noctuidae and Geometridae, and the wooly apple aphid, *Eriosoma lanigerum* (Hausmann, 1802) (Hemiptera: Aphididae), should be monitored continuously and additional control measures implemented as needed.

The integrated management of these pests associated with pheromones is essential for the implementation of integrated fruit production in Brazilian conditions.

Acknowledgements

To "Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq)" and "Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES)", ISCA Technologies Ltda. for the kind supply of sex pheromones and traps, "Embrapa Uva e Vinho" and "RASIP AGRO PASTORIL S.A.", Vacaria, Rio Grande do Sul State, for allowing the use of the company's orchard for the execution of this work.

Literature cited

ALBAJES, R.; KONSTANTOPOULOU, M.; ETCHEPARE, O.; EIZAGUIRRE, M.; FRÉROT, B.; SANS, A.; KROKOS, F.; AMÉLINE, A.; MAZOMENOS, B. 2002. Mating disruption of the corn borer *Sesamia nonagrioides* (Lepidoptera: Noctuidae) using sprayable formulations of pheromone. Crop Protection 21 (3): 217-225.

ANGELI, G.; BERTI, M.; MAINES, R.; IORIATTI, C. 2003. Utilizzo delle tecniche di disorientamento e atract kill nella difesa del mello da *Cydia pomonella* (L.) e *Cydia molesta* (Busck). Informatore Fitopatologico 9 (1): 45-50.

ARIOLI, C. J.; BOTTON, M.; CARVALHO, G. A. 2004. Controle químico da *Grapholita molesta* (Busck) (Lepidoptera: Tortricidae) na cultura do pessegueiro. Ciencia Rural 34 (6): 1695-1700.

ARIOLI, C. J.; ZART, M.; GARCIA, M. S.; BOTTON, M. 2007. Avaliação de inseticidas neonicotinóides para o controle da mariposa-oriental *Grapholita molesta* (Busck) (Lepidoptera: Tortricidae) em laboratório e pomar comercial de maçã com infestações artificiais. BioAssay 2 (1): 1-6.

ATANASSOV, A.; SHEARER, P. W.; HAMILTON, G. C. 2003**.** Peach pest management programs impact beneficial fauna abundance and *Grapholita molesta* (Lepidoptera: Tortricidae) egg parasitism and predation. Environmental Entomology 32 (4): 780-788.

BOSA, F.; OSORIO, P.; COTES, A. M.; BENGTSSON, M.; WITZGALL, P.; FUKUMOTO, T. 2008. Controle de *Tecia solanivora* (Lepidoptera: Gelechiidae) mediante su feromona para la interrupción del apareamiento. Revista Colombiana de Entomología 34 (1): 68-75.

BOTTON, M.; NAKANO, O.; KOVALESKI, A. 2000a. Controle químico da lagarta-enroladeira *Bonagota cranaodes* (Meyrick) na cultura da macieira. Pesquisa Agropecuária Brasileira 35 (11): 2139-2144.

BOTTON, M.; NAKANO, O.; KOVALESKI, A. 2000b. Exigências térmicas e estimativa do número de gerações de *Bonagota cranaodes* (Meyrick) (Lepidoptera: Tortricidae) em regiões produtoras de maçã do sul do Brasil. Anais da Sociedade Entomológica do Brasil 29 (4): 633-637.

BOTTON, M.; KULCHESKI, F.; COLLETTA, V. D.; ARIOLI, C. J.; PASTORI, P. L. 2005**.** Avaliação do uso do feromônio de confundimento no controle da *Grapholita molesta* (Lepidoptera: Tortricidae) em pomares de pessegueiro. Idesia 23 (1): 43-50.

BOTTON, M.; ARIOLI, C. J.; RINGENBERG, R.; MORANDI FILHO, W. J. 2009. Controle químico de *Bonagota salubricola* (Meyrick, 1937) (Lepidoptera: Tortricidae) em laboratório e pomar de macieira. Revista Arquivos do Instituto Biológico 76 (2): 225-231.

CARDÉ, R. T.; MINKS, A. T. 1995. Control of moth pests by mating disruption: Successes and constraints. Annual Review of Entomology 40 (1): 559-585.

CHARMILLOT, P. J.; VICKERS, R. A. 1991. Use of sex pheromones for control of tortricid pests in pome and stone fruits. pp. 487-496. In: Van Der Gueest, L. P. S.; Evenhuis, H. H. (Eds.). Tortricid pests, their biology, natural enemies and control. Elsevier Science. Amsterdam. 808 p.

CORACINI, D. A.; BENGTSSON, M.; RECKZIEGEL, A.; LOFQVIST, J.; FRANCKE, W.; VILELA, E. F.; EIRAS, A. E.; KOVALESKI, A.; WITZGALL, P. 2001. Identification of a four-component sex pheromone blend in *Bonagota cranaodes* (Lepidoptera: Tortricidae). Journal of Economic Entomology 94 (4): 911-914.

CRAVEDI, P.; GUARINO, F.; TOCCI, A. 2001. Valuations about mating disruption method application in *Cydia molesta* (Busck) control on nearly 400 hectares of peach tree in the Plane of Sibari (Calabria, South Italy). IOBC/WPRS Bulletin 24 (5): 79-84.

DEGEN, T.; CHEVALLIER, A.; FISCHER, S. 2005. Evolution de la lutte phéromonale contre les vers de la grape. Revue Suisse de Viticulture, Arboriculture and Horticulture 37 (5): 273-280.

EPSTEIN, D. L.; STELINSKI, L. L.; REED, T. P.; MILLER, J. R.; GUT, L. J. 2006. Higher densities of distributed pheromone sources provide disruption of codling moth (Lepidoptera: Tortricidae) superior to that of lower densities of clumped sources. Journal of Economic Entomology 99 (4): 1327-1333.

IL'ICHEV, A. L.; WILLIANS, D. G.; MILNER, A. D. 2004. Mating disruption barriers in pome fruit for improved control of oriental fruit moth, *Grapholita molesta* Busck (Lep., Tortricidae) in stone fruit under mating disruption. Journal of Applied Entomology 128 (2): 126-132.

JENKINS, J. W. 2002. Use of mating disruption in cotton in North and South America. IOBC/WPRS Bulletin 25 (9): 21-26.

JENKINS, P. E.; ISAACS, R. 2008. Mating disruption of *Paralobesia viteana* in vineyards using pheromone deployed in SPLAT-GBM™ wax droplets. Journal of Chemical Ecology 34 (8): 1089-1095.

JEANNERET, P.; CHARMILLOT, P. J. 1995. Movements of Tortricid moths (Lep. Tortricidae) between apple orchards and adjacent ecosystems. Agriculture, Ecosystems & Environment 55 (1): 37-49.

KARLSON, P.; LUSCHER, M. 1959**.** Pheromones, a new term for a class of biologically active substances. Nature 183 (1): 55-56.

KNIGHT, A. L.; LARSEN, T. E.; KETNER, K. C. 2004. Rainfastness of a microencapsulated sex pheromone formulation for codling moth (Lepidoptera: Tortricidae). Journal of Economic Entomology 97 (6): 1987-1992.

KOVALESKI, A.; BOTTON, M.; NAKANO, O.; VILELA, E. F.; EIRAS, A. E. 2003. Concentração e tempo de liberação do feromônio sexual sintético de *Bonagota cranaodes* (Meyrick) (Lepidoptera: Tortricidae) na cultura da macieira. Neotropical Entomology 32 (1): 45-48.

KOVANCI, O. B.; SCHAL, C.; WALGENBACH, J. F.; KENNEDY, G. G. 2005**.** Comparison of mating disruption with pesticides for management of oriental fruit moth (Lepidoptera: Tortricidae) in North Carolina apple orchards. Journal of Economic Entomology 98 (4): 1248-1258.

MAFRA-NETO, A.; ARIOLI, C. J.; BORGES, R. 2008. Feromônios: Futuro promissor. Revista Cultivar Hortaliças e Frutas 47 (1): 02-07.

MEISSNER, H. E.; WALGENBACH, J. F.; KENNEDY, G. G. 2001. Effects of mating disruption and conventional pesticide treatments on populations of the tufted apple bud moth, *Platynota idaeusalis*, in North Carolina apple orchards. Crop Protection 20 (5): 373-378.

MICHEREFF FILHO, M.; VILELA, E. F.; JHAM, G. N.; ATTYGALLE, A.; SVATOS, A.; MEINWALD, J. 2000. Initial studies of mating disruption of the tomato moth, *Tuta absoluta* (Lepidoptera: Gelechiidae) using synthetic sex pheromone. Journal of Brazilian Chemical Society 11 (6): 621-628.

MILLER, J. R.; GUT, L. J.; LAME, F. M.; STELINSKI, L. L. 2006a. Differentiation of competitive vs. non-competitive mechanisms mediating disruption of moth sexual communication by point sources of sex pheromone (Part I): Theory. Journal of Chemical Ecology 32 (10): 2089-2114.

MILLER, J. R.; GUT, L. J.; LAME, F. M.; STELINSKI, L. L. 2006b. Differentiation of competitive vs. non-competitive mechanisms mediating disruption of moth sexual communication by point sources of sex pheromone (Part II): Case studies. Journal of Chemical Ecology 32 (10): 2115-2143.

MOLINARI, F. 2002. Criteri per l'applicazione del metodo della confusione nella defesa del pesco. Notiziario Sulla Protezione Delle Piante 14 (1): 165-169.

MOLINARI, F.; CRAVEDI, P.; RAMA, F.; REGGIORI, F.; DAL PANE, M.; GALASSI, T. 2000. L'uso dei feromoni secondo il metodo del "disorientamento" nella difesa del pesco da *Cydia molesta* e *Anarsia lineatella*. Atti delle Giornate Fitopatologiche 1 (1): 341-348.

MOLINARI, F.; RAMA, F.; REGGIORI, F.; ZANREI, O. 2004. Evolution of the dispensers of pheromone for false-trail-following in the control of *Cydia molesta* (Busck) and *Anarsia lineatella* Zeller. IOBC/WPRS Bulletin 27 (5): 57-62.

MONTEIRO, L. B.; SOUZA, A.; BELLI, L. 2008. Confusão sexual para o controle de *Grapholita molesta* (Lepidoptera: Tortricidae), em pomares de macieira, em Fraiburgo (SC), Brasil. Bragantia 67 (1): 191-196.

MOSCHOS, T.; SOULIOTIS, C.; BROUMAS, T.; KAPOTHANASSI, V. 2004. Control of the European grapevine moth *Lobesia botrana* in Greece by the mating disruption technique: A three-year survey. Phytoparasitica 32 (1): 83-96.

PASTORI, P. L.; MONTEIRO, L. B.; BOTTON, M.; PRATISSOLI, D. 2007. Capacidade de parasitismo de *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae) em ovos de *Bonagota salubricola* (Meyrick) (Lepidoptera: Tortricidae) sob diferentes temperaturas. Neotropical Entomology 36 (6): 926-931.

PASTORI, P.L.; ARIOLI, C. J.; BOTTON, M.; MONTEIRO, L. B.; MAFRA-NETO, A. 2008. Avaliação da técnica de disrupção sexual utilizando emissores SPLAT® visando ao controle de *Bonagota salubricola* (Meyrick) e *Grapholita molesta* (Busck) (Lepidoptera: Tortricidae) na pré-colheita de maçãs da cultivar 'Fuji'. BioAssay 3 (1): 1-8.

PROTAS, J. F. S.; SANHUEZA, R. M. V. 2002. Normas técnicas e documentos de acompanhamento da produção integrada de maçã. Embrapa Uva e Vinho (Documentos, 33), Bento Gonçalves. 64 p.

RICCIOLINI, M.; BALDI, J. 1990. L'uso dei feromoni sessuali nella lotta alla Cydia del pesco (*Grapholita molesta* Busk.) col metodo della confusione. Un ulteriore passo avanti nella riduzione dell'impiego dei fitofarmaci nella difesa del frutteto. Agricoltura Toscana 5/6 (1): 21-24.

SALLES, L. A. B.; MARINI, L. H. 1989. Avaliação de uma formulação de confundimento no controle de *Grapholita molesta*

(Busck, 1916) (Lepidoptera: Tortricidae). Anais da Sociedade Entomológica do Brasil 18 (2): 329-336.

SIMON, S.; DEFRANCE, H.; SAUPHANOR, B. 2007. Effect of codling moth management on orchard arthropods. Agriculture, Ecosystems & Environment 122 (3): 340-348.

STELINSKI, L. L.; GUT, L. J.; MALLINGER, R. E.; EPSTEIN, D. L.; REED, T. P.; MILLER, J. R. 2005. Small plot trials documenting effective mating disruption of oriental fruit moth by using high densities of wax-drop pheromone dispensers. Journal of Economic Entomology 98 (4): 1267-1274.

STELINSKI, L. L.; MILLER, J. R.; LEDEBUHR, R.; GUT, L. J. 2006. Mechanized applicator for large scale field deployment of paraffin wax dispensers of pheromone for mating disruption in tree fruit. Journal of Economic Entomology 99 (5): 1705-1710.

STELINSKI, L. L.; MILLER, J. R.; LEDEBUHR, R.; SIEGERT, P.; GUT, L. J. 2007. Season-long mating disruption of *Grapholita molesta* (Lepidoptera: Tortricidae) by one machine application of pheromone in wax drops (SPLAT-OFM). Journal of Pest Science 80 (2): 109-117.

SUCKLING, D. M.; WALKER, J. T. S.; WEARING, C. H. 1999. Ecological impact of three pest management systems in New Zealand apple orchards. Agriculture, Ecosystems & Environment 73 (2): 129-140.

THOMSON, D.; BRUNNER, J.; GUT, L. J.; JUDD, G.; KNIGHT, A. 2001. Ten years implementing codling moth mating disruption in the orchards of Washington and British Columbia: Starting right and managing for success. IOBC/WPRS Bulletin 24 (2): 23-30.

TRIMBLE, R. M.; PREE, D. J.; CARTER, N. J. 2001. Integrated control of oriental fruit moth (Lepidoptera: Tortricidae) in peach orchards using insecticide and mating disruption. Journal of Economic Entomology 94 (2): 476-485.

UNELIUS, C. R.; EIRAS, A. E.; WITZGALL, P.; BENGTSSON, M.; KOVALESKI, A.; VILELA, E. F.; KARLSON, A. K. B. 1996. Identification and synthesis of the sex pheromone of *Phtheochroa cranaodes* Meyrick (Lepidoptera: Tortricidae). Tetrahedron Letters 37 (9): 1505-1508.

VICKERS, R. A.; ROTHSCHILD, G. H. L.; JONES, E. L. 1985. Control of oriental fruit moth *Cydia molesta* (Busck) (Lepidoptera: Tortricidae) at a district level by mating disruption with synthetic female pheromones. Bulletin of Entomological Research 75 (4): 625-634.

Received: 20-Jan-2011 • Accepted: 15-Nov-2012