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Communication interference in sympatrically occurring moth species

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Abstract

In moth species, females emit a species-specific sex pheromone that is perceived over long distance by conspecific males. The species-specificity in the chemical communication channel is achieved by a combination of unique components in specific ratios and sometimes also by interspecific behavioural antagonists to deter sympatrically occurring heterospecific males. In this study, we determined possible antagonistic effects in *Helicoverpa gelotopoeon* Dyar (Lepidoptera: Noctuidae) males to the major sex pheromone component of sympatrically occurring heliothine moths, Z11-16:Ald, as well as to the sex pheromone of the sympatrically occurring *Heliothis virescens* (Fabricius) (Lepidoptera: Noctuidae) (Z11-16:Ald and Z9-14:Ald). We also explored whether other co-occurring species are attracted to these pheromone blends. Our field experiments showed that the addition of Z11-16:Ald alone or in combination with Z9-14:Ald inhibited trap catches of *H. gelotopoeon* males and that this inhibition depended on the concentration of these compounds. In addition, other moth species were attracted to the blends. Together, our results confirm the antagonistic effect of heterospecific sex pheromone compounds of *H. virescens* to *H. gelotopoeon*.

Introduction

In moth sexual communication, females emit a species-specific sex pheromone blend that attracts conspecific males from a distance (Wyatt, 2003; Cardé & Haynes, 2004). The species-specificity is determined by a combination of the components and their relative amounts (Cardé & Haynes, 2004; Symonds & Elgar, 2008). In closely related species, sex pheromones may contain the same pheromone components, albeit in different proportions (Cardé & Haynes, 2004). This overlap in components generates the chance of communication interference and even cross-matings (Mitchell, 1976; Evenden et al., 1999; Symonds & Elgar, 2008). To avoid heterospecific attraction, the pheromone blend may also contain inhibitory compounds, which are known as antagonists (Cossé et al., 1998; Quero & Baker, 1999; Gemeno et al., 2006; Eizaguirre et al., 2007). These antagonistic pheromone compounds thus play a role in maintaining reproductive isolation between closely related species that coexist and have overlapping pheromone blends (Fadamiro & Baker, 1997; Vickers & Baker, 1997; Cardé & Haynes, 2004; Lelito et al., 2008).

South America has ca. 100 species of noctuid moths (Lepidoptera: Noctuidae), many of which have overlapping geographic distributions and share at least part of their host plant range (Pastrana et al., 2004). Particularly in Argentina, among the pests that cause economic

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losses, four species of heliothines co-occur: *Heliothis virescens* (Fabricius), *Helicoverpa zea* (Boddie), *Helicoverpa gelotopoeon* Dyar, and the recently introduced *Helicoverpa armigera* (Hübner). All four species are generalists, with a wide host range, including tobacco (*Nicotiana tabacum* L.), maize (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), soybean (*Glycine max* L. Merr.), chickpea (*Cicer arietinum* L.), alfalfa (*Medicago sativa* L.), and bean (*Phaseolus vulgaris* L.), among other crops (Reed & Pawar, 1982; Fitt, 1989; Cork & Lobos, 2003; Mastrangelo et al., 2014). In addition, the specialist *Heliothis subflexa* (Guenée) occurs when *Physalis* spec. plants are present (Bado et al., 2005).

In South American heliothine moths, (*Z*)-11-hexadecenal (Z11-16:Ald) is the major sex pheromone component (Roelofs et al., 1974; Vickers et al., 1991; Groot et al., 2005), except *H. gelotopoeon*, which has (*Z*)-9-hexadecenal (Z9-16:Ald) as the major pheromone component (Cork & Lobos, 2003). The species-specificity of the pheromone blend in *H. virescens*, *H. zea*, and *H. armigera* is due to the relative amount of minor components (Table 1). For example, in *H. virescens* the minor component (*Z*)-9-tetradecenal (Z9-14:Ald) is critical for the attraction of conspecific males (Roelofs et al., 1974; Teal et al., 1986), whereas in *H. zea* and *H. armigera* it is the addition of Z9-16:Ald in different proportions (Nesbitt et al., 1980; Pope et al., 1984). In *H. gelotopoeon*, hexadecanal (16:Ald) is the secondary critical sex pheromone component (Table 1). Interestingly, Z11-16:Ald is absent in the female pheromone blend of this species (Cork & Lobos, 2003).

Behavioural antagonism to pheromone compounds has been reported in these five heliothine species (Table 1). For example, the addition of low amounts of Z9-14:Ald (the secondary component in *H. virescens*) in a *H. zea* pheromone blend significantly reduced the captures of *H. zea* males (Shaver et al., 1982). In addition, traps with *H. virescens* and *H. zea* females placed together reduced the captures of *H. zea* males (Haile et al., 1973; Lopez & Witz, 1988). Also, the addition of Z11-16:OAc and Z11-16:OH, present in the pheromone of *H. subflexa*, significantly inhibited the attraction of *H. zea* males (Fadamiro & Baker, 1997; Lelito et al., 2008) and *H. virescens* males (Vickers & Baker, 1997; Groot et al., 2006; Lelito et al., 2008). In *H. armigera*, Z9-14:Ald and Z11-16:OH have also been reported to elicit an inhibitory response (Kehat et al., 1980; Kehat & Dunkelblum, 1990). In *H. gelotopoeon*, an inhibitory effect was found for Z11-16:Ald (Cork & Lobos, 2003).

As antagonistic behaviour has evolutionary significance and a possible practical application as a pest management tool, we conducted field experiments to compare the response of *H. gelotopoeon* males in the presence of Z11-16:Ald alone or in combination with Z9-14:Ald, the critical secondary sex pheromone component of *H. virescens*. In addition, we explored whether other co-occurring species were attracted to the various blends.

Materials and methods

Experimental site and general experimental procedures

Trapping experiments were carried out in two commercial
soybean fields near El Timbó (26°41.841′S, 65°06.834′W) and Las Cejas (26°52.428′S, 64°44.872′W), Tucumán province, northwest Argentina. The experiments were run during the summer season over a period of 3 months (January – March 2014). In each field site, we set up plots and each plot was used for a given experiment. Plots were spaced 30 m apart and consisted of three linear arrangements of traps placed 15 m apart. Inside each linear arrangement, we placed one trap per treatment (i.e., three replicates per treatment). Traps were hung 1.5 m above ground level, and trap position in each linear arrange-

Table 1 Sex pheromone components of co-occurring heliothine species in South America

Component	*Heliothis virescens*	*Helicoverpa zea*	*Helicoverpa armigera*	*Helicoverpa gelotopoeon*	*Heliothis subflexa*
Z11-16:Ald	++++	++++	++++	Antagonist	++++
Z9-14:Ald	+++	Antagonist	Antagonist		
16:Ald		++		++++	
14:Ald				++	
Z7-16:Ald		++			
Z9-16:Ald		+++	+++	++++	+++
Z11-16:OH	Antagonist	Antagonist	Antagonist	Antagonist	+++
Z11-16:OAc	Antagonist	Antagonist			

++++: major sex pheromone component, +++: critical secondary sex pheromone component, ++: minor sex pheromone component, and antagonist: component that avoids attraction between heterospecifics.

ment within a plot was randomized. In all trapping experiments, the synthetic pheromone blends were placed in locally produced traps (Huber & Hoffmann, 1979). Specifically, traps consisted of 1-l plastic buckets with four equally spaced holes each (5 cm diameter) drilled through the vertical wall, 2 cm below the lid. The buckets contained water with a thin layer of light motor oil to kill the captured males. The septa were fixed by the wire to the underside of lids of traps. Every 2–3 days, trapped moths were collected from the traps, after which the traps were rotated to avoid position effects. Experiments finished when all treatments were permuted over all possible positions within each linear arrangement. The collected males were stored either at 8 °C or in 70% alcohol for species identification.

Male trapping experiments

To determine the effect on the response of *H. gelotopoeon* males and other species to the addition of Z11-16:Ald and Z9-14:Ald to *H. gelotopoeon* pheromone, four dose-response experiments were performed (see Table 2). (1) Response of males to high concentrations of Z11-16:Ald: either 10, 50, or 100% of this component was added to the *H. gelotopoeon* pheromone blend, which consisted of Z9-16:Ald, 16:Ald, and 14:Ald (referred to as *Hg* blend). (2) Response of males to low concentrations of Z11-16:Ald; either 1 or 10% of this component was added to the *Hg* blend. (3) Change in the response as a result of the addition of the two critical sex pheromone components of *H. virescens* pheromone, Z11-16:Ald and Z9-14:Ald (referred to as *Hv* blend); either 1, 10, 50, or 100% of this blend was added to the *Hg* blend. (4) Specific comparison of the response of *H. gelotopoeon* males to the *Hg* vs. *Hg* blend with 1% *Hv* blend, to verify our results of experiment 3 when adding the *Hv* to the *Hg* blend at the lowest dose (1%).

Preparation of pheromone lures

Pheromone compounds used to prepare the lures were purchased from Pherobank (Wageningen, The Netherlands). The treatment solutions were prepared in hexane and contained the major pheromone component of *H. gelotopoeon* (Z9-16:Ald) plus the corresponding amounts of the other two components, 16:Ald and 14:Ald, in the proportions reported by Cork & Lobos (2003). In addition, and depending on the treatment, the blends also contained different amounts of Z11-16:Ald (experiments 1 and 2) or Z11-16:Ald and Z9-14:Ald (experiments 3 and 4) in the respective proportions (see Table 2). Butylated hydroxytoluene (BHT, 1%) was added to avoid degradation of the compounds. Red rubber septa (Pherobank) were soaked in hexane for 24 h, air dried for 3-4 h, and stored until used. Each septum received 100 µl of the treatment solutions and contained 100 µg of Z9-16:Ald, with all the other components in the corresponding amounts. To confirm the proportions of each compound, each solution was checked on a gas chromatograph (GC) before they were loaded onto the septa. After the addition of the pheromone blend, the septa were dried for 40 min, wrapped in aluminium foil, placed in plastic bags, and preserved at −20 °C. Control traps contained septa soaked only in hexane.

Chemical analysis

To verify the purity and composition of the treatment solutions, GC analysis was performed at the Departamento de Química Aplicada y Alimentos, Facultad de Agronomía, Universidad de Buenos Aires, Argentina, using an Agilent 7890A equipped with a HP-5 column (30 m × 0.32 mm i.d. × 0.25 µm film thickness; Agilent Technologies, Wilmington, DE, USA), and a flame ionization detector (FID). The oven temperature was programmed from 60 °C (held for 2 min) to 180 °C at 15 °C per min, then to 230 °C at 5 °C per min, and to 245 °C at 20 °C min and then held for 10 min. Samples were injected in the splitless mode with the injector purged at 30 s with nitrogen as the carrier gas at 27.6 cm s^{-1} flow velocity.

Table 2 Amount (µg) of Z11-16:Ald and Z9-14:Ald (*Heliothis virescens* sex pheromone components) added to *Helicoverpa gelotopoeon* pheromone blends (16:Ald, 14:Ald, and Z9-16:Ald) used in the various experiments

	Treatment[1,2]					
Component	*Hg* blend	1%	10%	50%	100%	Control[3]
16:Ald	100	100	100	100	100	
14:Ald	2	2	2	2	2	
Z9-16:Ald	100	100	100	100	100	
Z11-16:Ald[1,2]	–	1	10	50	100	
Z9-14:Ald[2]	–	0.05	0.50	2.50	5	

[1]In experiments 1 and 2, the addition of 1–100% refers to the addition of Z11-16:Ald.
[2]In experiments 3 and 4, the addition of 1–100% refers to the addition of Z11-16:Ald and Z9-14:Ald.
[3]The rubber septa were soaked with the solvent hexane only.

Species identification

The species captured in all traps were identified using diagnostic characters of the male genitalia (Hardwick, 1965; Pastrana et al., 2004). Individuals were placed in a Petri dish and, with the aid of fine forceps, the genitalia were dissected from the abdomen and the aedeagus was removed. The aedeagus was then everted, which allowed proper identification. Voucher samples were deposited in the laboratory of Cátedra de Terapéutica Vegetal, Facultad de Agronomía y Zootecnia, Universidad Nacional de Tucumán, Argentina.

Statistical analysis

Data of all experiments were analysed using InfoStat and R software (Di Rienzo et al., 2012; R Core Team, 2015). Each species and site was analysed separately and all treatments, except the control (hexane) with which we did not catch any males, were included in the statistical analysis. For experiments 1–3, the trap catches were log transformed to stabilize the variance. To determine the dose effect of Z11-16:Ald (experiments 1 and 2) or the *Hv* blend (experiments 3) on the number of *H. gelotopoeon* males caught per trap, different mixed effect regression models were explored and the model with the least mean square error was chosen. The fixed factor was dose, whereas the random factor was the combination of the linear arrangement and rotation. Experiment 4 was analysed using generalized linear mixed models (GLMM) with Poisson error distribution and log link function using the lme4 package from R (Bates et al., 2014). Dose and site were the fixed factors and the random factor was the combination of rotation and linear arrangement within each site. Least significant difference (LSD) test with Sidak's correction for multiple comparisons was used to compare means among doses (Bretz et al., 2001). To model trap catches of the other species, we used a non-linear regression for experiment 1 and GLMM for experiment 3.

Results

Helicoverpa gelotopoeon was captured in all experiments. Overall, the field experiments showed that the addition of Z11-16:Ald alone or in combination with Z9-14:Ald inhibited the catches of *H. gelotopoeon* males in a dose-dependent manner. Specifically, when we tested the effect of Z11-16:Ald to determine an inhibitory effect of *H. gelotopoeon* males (experiment 1), we found that Z11-16:Ald strongly reduced the response of *H. gelotopoeon* males: when adding 50 or 100% we hardly caught any males at all, whereas the addition of 10% already reduced the number of males caught from a mean (± SEM) of 56.7 ± 9.3 to 5 ± 2 males per trap in El Timbó and from 102.3 ± 22.7 to 2.3 ± 0.7 males per trap in Las Cejas. The trap catches fitted an exponential function (Figure 1A, Table 3).

When we evaluated lower doses (experiment 2), Z11-16:Ald also reduced the response of *H. gelotopoeon* males. These data fitted a linear function (Figure 1B, Table 3). The addition of 1% Z11-16:Ald reduced the average trap catches from 20.3 ± 2.9 to 8.7 ± 3.3 males per trap in El Timbó and from 120.3 ± 12.2 to 92.3 ± 5.9 males per trap in Las Cejas. In traps with 10% Z11-16:Ald, we caught 2.3 ± 0.9 and 21 ± 3.2 males per trap in El Timbó and Las Cejas, respectively.

When adding the *Hv* blend to the *Hg* blend (experiment 3), we also found a reduction in the trap captures. These data fitted an exponential function (Figure 1C, Table 3). The addition of 1% *Hv* blend reduced the trap

Table 3 Parameters for the regression models obtained to explain the relationship between *Helicoverpa gelotopoeon* and *Neotuerta platensis* males trapped when Z11-16:Ald was added to the *Hg* blend alone or in combination with Z9-14:Ald (*Hv* blend) at various doses in three experiments in soybean fields in El Timbó and Las Cejas, Tucumán, Argentina

Species	Experiment	Site	a	95% CI	b	95% CI	r^2
H. gelotopoeon	1	El Timbó	0.96	0.84–1.08	−0.14	−0.19–[−0.08]	0.71
		Las Cejas	1.23	1.13–1.33	−0.24	−0.32–[−0.16]	0.88
	2	El Timbó	0.71	0.61–0.81	−0.05	−0.07–[−0.03]	0.49
		Las Cejas	1.56	1.48–1.64	−0.07	−0.09–[−0.05]	0.77
	3	El Timbó	0.94	0.78–1.10	−0.31	−0.41–[−0.21]	0.74
		Las Cejas	1.2	0.98–1.42	−0.16	–0.28–[−0.04]	0.79
N. platensis	1	El Timbó	0.04	0.00–0.08	0.03	0.01–0.05	0.52
		Las Cejas	0.12	0.02–0.22	0.02	0.01–0.03	0.50

For experiments 1 and 3: $y = a \times e^{b \times [dose]}$; for experiment 2: $y = a + b \times [dose]$.
Experiment 1: *Hg* blend with the addition of Z11-16:Ald at 1 and 10%; Experiment 2: *Hg* blend with the addition of Z11-16:Ald at 10, 50, and 100%; Experiment 3: *Hg* blend with the addition of *Hv* blend at 1, 10, 50, and 100%.

catches from an average of 61.7 ± 8.4 to 36.3 ± 4.6 *H. gelotopoeon* males per trap in El Timbó and from 100 ± 6.7 to 91.7 ± 10.9 males per trap in Las Cejas. Trap captures with 10% *Hv* blend added to the *Hg* blend were 0.3 ± 0.3 and 4.7 ± 2.7 males per trap in El Timbó and Las Cejas, respectively. In traps to which 50% *Hv* blend was added, an average of 0.3 ± 0.3 and 1 ± 1 males per trap were caught in El Timbó and Las Cejas, respectively. With the addition of 100% *Hv* blend, we captured in total three males in Las Cejas and none in El Timbó.

When we added only the lowest dose of *Hv* blend to the *Hg* blend (experiment 4), in El Timbó we caught an average of 9.7 ± 3.5 males per trap baited with the *Hg* blend, compared to 3 ± 0.6 males per trap baited with *Hg* blend +1% *Hv* blend (Figure 1D). In Las Cejas, the number of males caught in the traps with 1% *Hv* blend was significantly reduced from 111.7 ± 4.4 to 32.7 ± 7.4 males per trap (Figure 1D).

In addition to *H. gelotopoeon*, we caught *H. virescens* males, specifically in traps with the *Hg* blend to which the *Hv* blend was added (experiment 3). In El Timbó, we

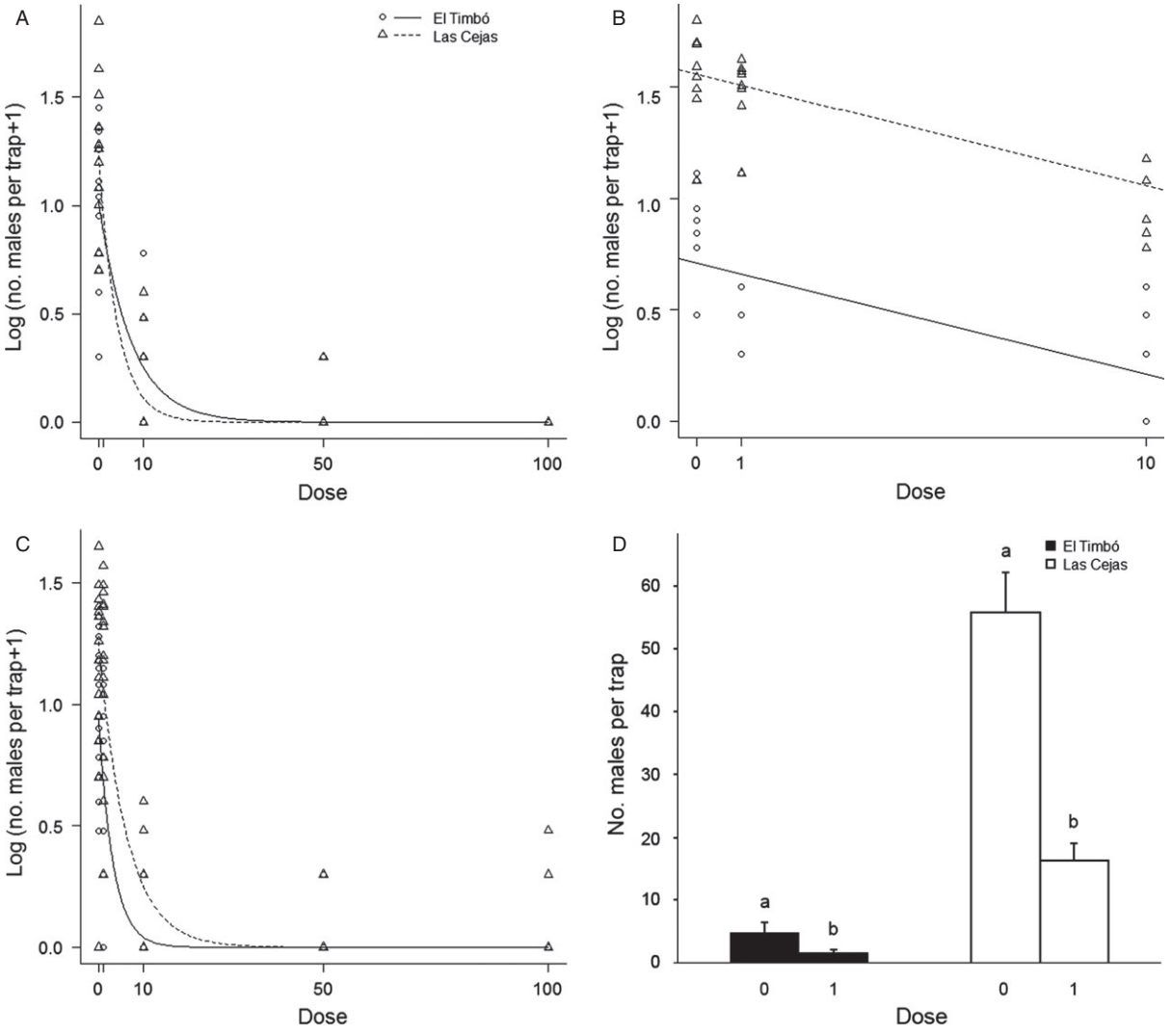

Figure 1 Number of *Helicoverpa gelotopoeon* males per trap in dose-response experiments with various concentrations of Z11-16:Ald and *Hv* blend added to *Hg* blend in soybean fields in El Timbó and Las Cejas, Tucumán, Argentina. (A) Experiment 1: addition of 0, 10, 50, or 100% Z11-16:Ald to *Hg* blend; (B) experiment 2: addition of 0, 1 or 10% Z11-16:Ald to *Hg* blend; (C) experiment 3: addition of 0, 1, 10, 50, or 100% *Hv* blend to *Hg* blend; (D) mean (+ SE) number of males per trap after addition of 0 or 1% *Hv* blend to *Hg* blend. The lines in A-C correspond to the mixed effect regression models. Data analysis in D is based on generalized linear mixed models (GLMM): means within a location capped with different letters are significantly different ($P<0.05$).

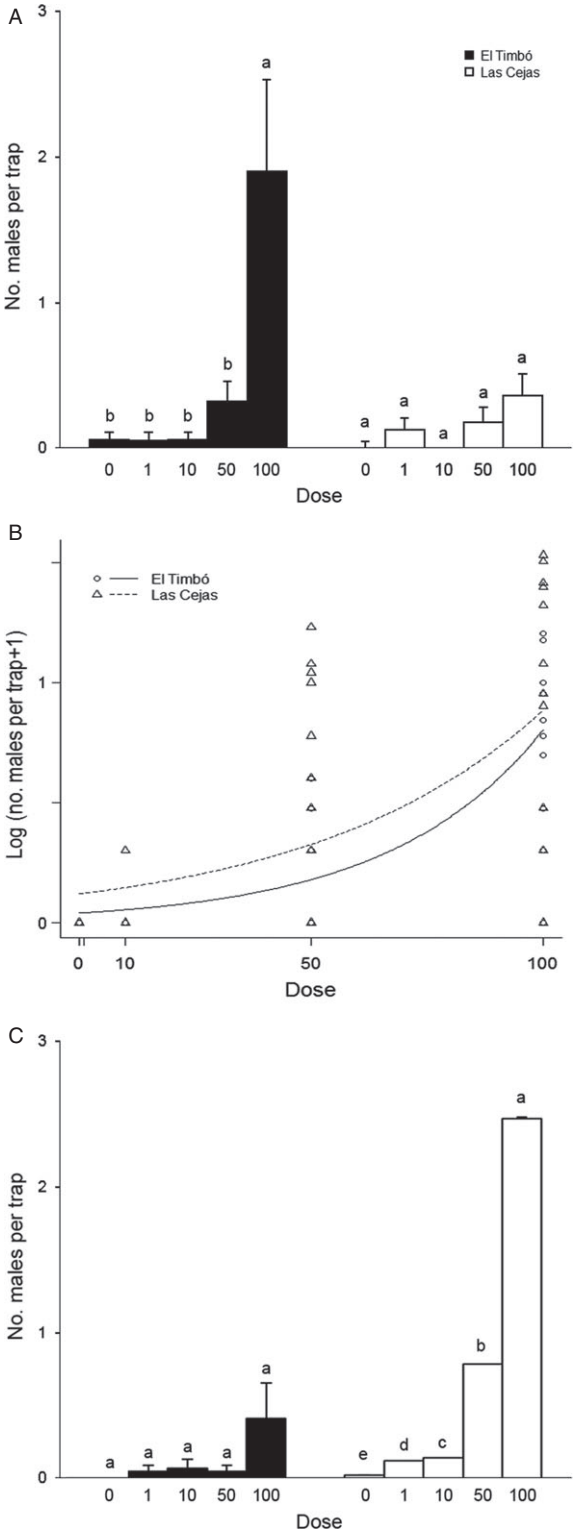

Figure 2 Number of other moth species males per trap in dose-response experiments with various concentrations of Z11-16:Ald and *Hv* blend added to *Hg* blend in soybean fields in El Timbó and Las Cejas, Tucumán, Argentina. (A) Mean (+ SE) number of *Heliothis virescens* males per trap in experiment 3: 0, 1, 10, 50, or 100% *Hv* blend to *Hg* blend; (B) number of *Neotuerta platensis* males per trap in experiment 1: addition of 0, 10, 50, or 100% Z11-16:Ald to *Hg* blend.; (C) mean (+ SE) number of *N. platensis* males in experiment 3. In A and C, means within a location capped with the same letter are not significantly different ($P>0.05$). In B, the lines correspond to the mixed effect regression models.

found a significant dose-response effect (Figure 2A): in traps with no *Hv* blend added to the *Hg* blend, we captured an average of 0.7 ± 0.3, in traps with the addition of 1 or 10% *Hv* blend we captured 0.7 ± 0.7 *H. virescens* males per trap, in traps with 50% *Hv* blend added to the *Hg* blend we caught 3.3 ± 0.9 males, whereas the addition of 100% *Hv* blend resulted in a trap catch of 19.7 ± 4.3 *H. virescens* males per trap. In Las Cejas, we caught much fewer *H. virescens* males (Figure 2A): in traps with no *Hv* blend added to the *Hg* blend, we captured no males, in traps with 1% *Hv* blend, we caught 1 ± 0.6 males per trap, in traps with 10% *Hv* blend we did not catch any *H. virescens* males, in traps with 50% *Hv* blend added to the *Hg* blend we caught 1.3 ± 0.9 males per trap, and in traps with 100% *Hv* blend we caught 2.7 ± 0.7 *H. virescens* males per trap.

Besides *H. gelotopoeon* and *H. virescens*, we also caught *Neotuerta platensis* (Berg) (Lepidoptera: Noctuidae) males at relatively high numbers (Figure 2B and C). Most captures occurred in the *Hg* blend to which 100% Z11-16:Ald was added alone or in combination with Z9-14:Ald (Figure 2B and C). For experiment 1, in El Timbó, we did not catch any individuals in traps without Z11-16:Ald added to the *Hg* blend or in traps to which 10% Z11-16:Ald was added. In traps with 50% Z11-16:Ald we caught an average of 4.3 ± 0.9 males per trap and in traps with 100% Z11-16:Ald we caught 25 ± 3.8 *N. platensis* males per trap. In Las Cejas, we captured no males in traps without Z11-16:Ald added to the *Hg* blend, whereas we caught 0.3 ± 0.3 males per trap with 10% Z11-16:Ald, 21.3 ± 3.8 males per trap with 50% Z11-16:Ald, and 67.7 ± 2.3 males per trap with 100% Z11-16:Ald. These data fitted an exponential function (Figure 2B, Table 3). For experiment 3, in El Timbó we captured no *N. platensis* males in traps without *Hv* blend added to the *Hg* blend; when 1% *Hv* blend was added to the *Hg* blend we captured an average of 0.7 ± 0.7 males per trap, in traps with 10% *Hv* blend we caught 1 ± 0.6 males per trap, in traps with 50% *Hv* blend we caught 0.7 ± 0.3 males per trap and in traps to which 100% *Hv* blend was added we caught 5.7 ± 4.2 males per trap. In Las Cejas, the GLMM analysis revealed a significant dose-response effect (Figure 2C): in traps with no *Hv*

blend added to the *Hg* blend, we captured an average of 0.3 ± 0.3 *N. platensis* males per trap, in traps with 1% *Hv* blend we caught 2 ± 2 males per trap, in traps with 10% *Hv* blend we caught 2.3 ± 1.2 males per trap, in traps with 50% *Hv* blend we caught 12.7 ± 2.6 males per trap, and in traps with 100% *Hv* blend we caught 40 ± 7.9 males per trap.

Discussion

Our field experiments revealed that both the major sex pheromone component of co-occurring heliothine species, Z11-16:Ald, as well as the *Hv* blend have an inhibitory effect on *H. gelotopoeon* males. In addition, other moth species responded in a dose-dependent way.

The compound Z11-16:Ald clearly acts as a strong antagonist for *H. gelotopoeon* males: significantly fewer *H. gelotopoeon* males were caught when Z11-16:Ald was present at 10%, whereas at higher doses we hardly caught any *H. gelotopoeon* males. Cork & Lobos (2003) reported a reduction in trap captures when Z11-16:Ald was present at 1%, although with some variability across the season. We also found variable results at this low dose, depending on the field site, which could be attributable to differences in abundance of *H. gelotopoeon*: in El Timbó, the number of *H. gelotopoeon* males caught was lower in all traps compared to the number of males captured in Las Cejas. Thus, it seems that 1% Z11-16:Ald is the response threshold at which the attraction of *H. gelotopoeon* males can already be inhibited. The addition of the sex pheromone blend of *H. virescens* (Z11-16:Ald and Z9-14:Ald) also elicited an inhibitory response in *H. gelotopoeon* males. Whether Z9-14:Ald alone has an inhibitory effect on *H. gelotopoeon* males remains to be tested. As *H. gelotopoeon* females do not produce any Z11-16:Ald, the antagonistic effect of Z11-16:Ald alone and in combination with Z9-14:Ald on trap catches of *H. gelotopoeon* males indicates that communication interference exists between *H. gelotopoeon* and other co-occurring heliothine moths. This interference could be exploited in pest management strategies, for example by saturating the air with Z11-16:Ald alone or with the pheromone blend of *H. virescens*, which would likely cause mating disruption in both *H. gelotopoeon* and *H. virescens*. Mating disruption is a successful pest management strategy used against other moth species (Witzgall et al., 2010).

Besides the inhibitory response that we found in *H. gelotopoeon* males, other co-occurring species responded to the blends tested. *Heliothis virescens* males were captured mostly in traps baited with *Hg* blend with the addition of 100% *Hv* blend. The fact that we only caught *H. virescens* males in traps baited with the *Hg* blend and 100% *Hv* blend confirms that *H. virescens* males are only attracted when Z9-14:Ald is added (Roelofs et al., 1974; Teal et al., 1986). In addition, we caught many *N. platensis* males in traps baited with the *Hg* blend and 50 or 100% of Z11-16:Ald, as well as in traps baited with the *Hg* blend and 50 or 100% of *Hv* blend. *Neotuerta platensis* is distributed in Argentina, Brazil, and Uruguay and associated mostly with plants from the families Fabaceae, Laureaceae, Portulacaceae, Cactaceae, and Vitaceae (Pastrana et al., 2004). Information regarding this species is scarce and to our knowledge there are no records on its impact as a crop pest, nor on its sex pheromone composition. Because traps with the *Hg* blend alone (Z9-16:Ald, 16:Ald, 14:Ald) did not catch *N. platensis* males, and the addition of Z9-14:Ald did not increase trap catches of *N. platensis*, it seems that Z11-16:Ald could be a compound involved in the response of *N. platensis* males. It would be interesting to analyse the chemical composition in the female sex pheromone gland and determine the response of *N. platensis* to traps baited only with Z11-16:Ald.

In summary, our results confirm the antagonistic effect of heterospecific sex pheromone compounds of *H. virescens* to *H. gelotopoeon* males. Because the use of antagonistic compounds to hamper the communication channels of insect pests has potential as a pest management tool, it would be useful to explore the use of Z11-16:Ald and Z9-14:Ald in mating disruption experiments. In addition, we found that *N. platensis* males were attracted to many pheromone blends that we used, especially blends containing as much *Hg* pheromone as Z11-16:Ald. This indicates that Z11-16:Ald could be involved in the response of *N. platensis* males.

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