



Wheat-pea intercropping for aphid control: from laboratory tritrophic approach to field application

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Abstract

Intercropping is an interesting practice to promote the sustainable control of insect pests such as aphids. In particular, volatile organic compounds emitted by aphid-infested intercropped plants may deter other aphid species from their host plants, while attracting natural enemies. In this study, olfactometer and net-cage behavioural assays were first conducted to determine the effect of wheat-pea mixtures combined with aphid infestations on odour preferences of the wheat aphid *Sitobion avenae* and two associated predator species, the ladybird *Harmonia axyridis* and the hoverfly *Episyrphus balteatus*. Healthy wheat plants were preferred by *S. avenae*, while wheat-pea mixtures combined with aphid infestations were significantly less attractive. *H. axyridis* preferred odours from healthy wheat plants mixed with aphid-infested pea plants. As for *E. balteatus*, their searching and oviposition behaviours were stimulated by the different wheat/pea combinations associated with aphid infestations. A field trial was also carried to compare the effect of mix and strip cropping wheat with pea on aphids and their natural enemies with both monocultures. Wheat and pea aphid populations were significantly reduced by both types of intercropping when compared to monocultures. Moreover, higher abundances of hoverflies, lacewings and ladybirds were found in wheat mixed with pea field, followed by strip cropping and monocultures. These findings show that wheat-pea intercropping can be efficient to reduce aphid populations, namely by promoting their biological control.

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Introduction

Annual monoculture cropping systems greatly simplified agroecosystems landscape structural diversity, favouring the establishment of pest populations (Andow, 1991; Landis and Marino, 1999). In order to reduce the use of insecticides, which have negative effects on human health (WHO, 1990) and environment (Devine and Furlong, 2007), alternative pest control methods have been developed, namely based on habitat management practices (Gurr *et al.*, 2004; Hassanali *et al.*, 2008). Among these, intercropping, which is considered as the cultivation of at least two plant species in the same place at the same time (Andrews and Kassam, 1976; Ofori and Stern, 1987; Anil *et al.*, 1998), can be interesting for the sustainable control of pests (Smith and McSorley, 2000; Hassanali *et al.*, 2008; Konar *et al.*, 2010; Suresh *et al.*, 2010; Vaiyapuri *et al.*, 2010). Focusing on pea (*Pisum sativum* Linnaeus)-wheat (*Triticum aestivum* Linnaeus) intercropping systems, beneficial effects were already observed on aphid control. In fact, this practice can significantly decrease pea aphid, *Acyrtosiphon pisum* (Harris) (Ndzana *et al.*, 2014; Lopes *et al.*, 2015), and English grain aphid, *Sitobion avenae* (Fabricius) (Zhou *et al.*, 2009a; Lopes *et al.*, 2015), populations. However, the mechanisms explaining how wheat-pea intercropping promotes aphid control, which is called associational resistance (Tahvanainen and Root, 1972), are not still well understood (Ndzana *et al.*, 2014).

The *resource concentration* hypothesis from Root (1973) states that phytophagous insects are more likely to find their host plants when those are concentrated in dense or pure stands. Increasing plant diversity by intercropping two or more plant species may affect the visual and olfactory location of herbivore's host plants, as reviewed by Poveda *et al.* (2008) and Barbosa *et al.* (2009). Focusing on chemical cues, host plants location may be disrupted when their odours are blended with neighboring non-host plants. As shown by Xie *et al.* (2012), winged *S. avenae* prefer wheat plant odours alone than blended odours of wheat intercropped with mung bean (*Vigna radiate* Linnaeus).

Moreover, herbivore-induced plant volatiles (HIPVs) emitted by aphid-infested non-host intercropped plants may deter other aphid species from their host plants. It is namely the case with methyl salicylate (MeSA), which can be emitted for example by aphid-infested hops (*Humulus lupulus* Linnaeus) (Campbell *et al.*, 1993) and soybean (*Glycine max* (Linnaeus) Merrill) (Zhu and Park, 2005) and repel cereal aphid species (Pettersson *et al.*, 1994). Moreover, HIPVs such as MeSA may also attract aphid natural enemies (Hatano *et al.*, 2008), such as the ladybird *Coccinella septempunctata* Linnaeus (Zhu and Park, 2005) and the hoverfly *Toxomerus marginatus* (Say) (Rodriguez-Saona *et al.*, 2011). However, the effect of HIPVs on aphids and their natural enemies has not been studied in the context of wheat-pea intercropping to our knowledge.

Therefore, the aim of this study was to determine the behavioural preferences of *S. avenae*, an important pest species that transmits efficiently the *Barley yellow dwarf virus* (BYDV) to wheat (Gray *et al.*, 1998), when exposed to blended odours of pea plants infested by the pea aphid, *A. pisum*, intercropped with healthy or *S. avenae* infested wheat plants. The same plant-aphid combinations were used to assess the behavioural preferences of two important aphid predator species, namely the multicolored Asian lady beetle, *Harmonia axyridis* (Pallas), and the marmalade hoverfly, *Episyrphus balteatus* DeGeer. Complementarily, a field trial was conducted to assess the effect of wheat-pea intercropping on aphids and their natural enemies in real environmental conditions.

Materials and methods

Plants and insects

Wheat (variety "Tybalt") and pea (variety "James") were sown in plastic pots (9 × 8 × 10 cm). After plant germination, *S. avenae* and *A. pisum* were transferred into wheat and pea respectively. Aphids were moved to newly emerged plants each week to guarantee their proper development.

H. axyridis adults were placed in aerated plastic boxes containing sugar, water-impregnated cotton, and multi-flower pollen. *E. balteatus* adults were reared in cages (75 × 60 × 90 cm) containing bee-collected pollen, sugar and water. Plants and insects were kept in a climate-controlled room (16:8 light/dark; 22 ± 1 °C).

S. avenae and *H. axyridis* olfactometer behavioural assays

A two-arm olfactometer similar to the one described by Vet *et al.* (1983) was used to test *S. avenae* and *H. axyridis* preferences for olfactory cues derived from wheat-pea mixtures combined with aphid infestations. The olfactometer was made entirely from Teflon and was closed with a removable glass roof. The walking arena was 40 cm wide (from center to odor source) and 1.5 cm high (from Teflon walking arena to glass ceiling). Charcoal-filtered air was pushed in each of the olfactometer arms through Teflon tubing and adjusted to 150 ml/min with a digital flowmeter. A pump ventilated the walking arena by removing air from the center at 300 ml/min. A 1-l glass chamber (inner diameter: 10 cm; height: 145 cm) was connected to one of the olfactometer arms and was used to dispose the odor source.

Eight dual choices were examined by comparing one of the following odour sources to clean air: (1) 20 healthy wheat plants, (2) 20 healthy pea plants, (3) 20 aphid-infested wheat plants (infested with 50 *S. avenae* 24 hours prior to the experiment), (4) 20 aphid-infested pea plants (infested with 50 *A. pisum* 24 hours prior to the experiment), (5) 20 healthy wheat plants mixed with 20 healthy pea plants, (6) 20 aphid-infested wheat plants (same conditions as above) mixed with 20 healthy pea plants, (7) 20 healthy wheat plants mixed with 20 aphid-infested pea plants (same conditions as above), (8) 20 aphid-infested wheat plants mixed with 20 aphid-infested pea plants (same conditions as above for both).

Forty winged *S. avenae* were individually placed in the center of the olfactometer. Their choice was recorded when they crossed a “choice line”, which was located 5 cm past the center of the walking arena, in direction of each odour sources.

Aphids that did not cross a line within 10 min were recorded as non-responders and excluded from analysis. Concerning *H. axyridis*, 20 females were individually randomly placed in the centre of the olfactometer. Their choice was determined by the time spent in each olfactometer zone. The duration of repetitions was fixed at three minutes, which was sufficient for individuals to explore the olfactometer arena. Those who did not cross a line within three minutes were considered as non-responders and excluded from analysis. Each aphid and lady beetle was tested only once. The olfactometer was cleaned with norvanol after each repetition. Experiments were conducted in a laboratory at 22 ± 1 °C and under uniform lighting.

E. balteatus behavioural observations

Visual observations were conducted in a controlled environment room (22 ± 1 °C). To do so, a net-cage (180 × 60 × 90 cm) (Fig. 1.) was set up in a black box (200 × 70 × 100 cm) consisting of a steel frame covered with black cardboard paper to avoid external visual cues. Uniform illumination was provided inside the box by four fluorescent light tubes (70 W; Luminux) positioned 10 cm above the net-cage.

Three pots containing wheat and pea plants were placed in each side of the net-cage as presented in Fig. 1. *E. balteatus* females were collected from rearing cages and individually placed in the center of the net-cage. Their behaviour was then recorded during 10 min using the Observer® software (Noldus information Technology, version 5.0, Wageningen, The Netherlands). Five behavioural events were observed as follows: (1) immobility: the hoverfly was immobilized on the cage without moving, (2) extensive flying: the hoverfly hovered in the cage far away the plant, (3) searching: the hoverfly hovered in the cage close to the plant, (4) acceptance: the hoverfly landed on the plant, stayed immobile or walked on it, with proboscis extension on the plant surface, (5) oviposition: the hoverfly female showed abdomen bending and laid eggs. Then individuals were tested for each treatment.

Twelve series of dual-choice experiments were compared (Table 1). The net cage was cleaned with norvanol after each test.

Field experimental design

To assess the effect of wheat-pea intercropping on aphids and their natural enemies, a field study was conducted in the experimental farm of Gembloux Agro-Bio Tech, University of Liege, Namur Province of Belgium (50°33'N, 4°42'E) in 2011.

The field trial consisted of four treatments: (1) wheat mixed with pea (WMP), (2) alternate strips of wheat and pea (SWP), (3) wheat monoculture (WM), (4) pea monoculture (PM). Plots positioned within wheat crops were settled by delimiting three distinct areas (4m × 10m each) for each treatment (total of 12 plots) (Fig. 2.). Wheat (variety "Tybalt") monoculture was planted in 20-cm-apart rows at a rate of 350 seeds per m² on 18 February 2011. Pea (variety "James") monoculture was planted in 50-cm-apart rows at a rate of 80 seeds per m² on 18 February in 2011. For wheat mixed with pea, pea was planted between the two rows of wheat at a rate of 35 seeds per m². No insecticide or herbicide was used in the whole experimental area. Wheat and pea were maintained with standard agronomic practices used in Europe.

Insect diversity and abundance monitoring

Yellow pan traps (Flora®, 27 cm diameter and 10 cm depth), which are frequently used to attract and trap insects (Laubertie *et al.*, 2006), were attached to fiberglass sticks and placed 10 cm above the surface of plants. Traps were filled with water and a few drops of detergent. A single trap was installed in the middle of each investigated plot (total of three traps per treatment). Traps were emptied and reset at 7-day intervals between 4 May and 29 June. Insects were collected and transferred to plastic 50-mL vials containing 70% ethanol. Aphids and their natural enemies were sorted and identified to the species level in the laboratory according to the following keys: Taylor (1981) for aphids, Roy *et al.* (2013) for ladybirds; van Veen (2010) for hoverflies; San Martin (2004) for lacewings. The number of individuals per species was also recorded.

Visual observations on plants were also performed to visually assess the diversity and abundance of aphids on wheat tillers and pea plants. To do so, 20 tillers or plants (both in intercropping treatments) were randomly observed in each plot.

Statistical analysis

Observed frequencies related to the choice of *S. avenae* and *H. axyridis* in olfactometer behavioural assays were compared to corresponding theoretical frequencies by using a χ^2 goodness-of-fit test. A Student's t test was performed to compare the mean frequencies of *E. balteatus* responses to wheat and pea stimuli. For field experiments, a data sqrt (n + 1) transformation was applied to stabilize the variance before each test. The density of insect populations was compared among treatments using a one-way analysis of variance (ANOVA), followed by Tukey's honestly significant differences (HSD) test. All statistical tests were performed using Minitab® 16.

Results

S. avenae and H. axyridis olfactometer behavioural assays

A strong preference of winged *S. avenae* was observed for healthy wheat ($\chi^2 = 32.00$, $P < 0.001$) and pea ($\chi^2 = 24.50$, $P < 0.001$) plants odours (Fig. 3.). However, *S. avenae* were not significantly attracted by odours from aphid-infested wheat plants and by aphid-infested wheat plants combined with aphid-infested pea plants. Significantly higher proportions of non-responding individuals were observed when exposed to odours from infested pea plants ($\chi^2 = 18.00$, $P < 0.001$), as well as with the other three combinations: wheat and pea ($\chi^2 = 24.50$, $P < 0.001$), wheat infested with aphids and pea ($\chi^2 = 4.50$, $P < 0.05$), wheat and pea infested with aphids ($\chi^2 = 12.50$, $P < 0.001$).

Proportionally, *H. axyridis* females spent significantly more time on aphid-infested wheat plant odours ($\chi^2 = 7.50$, $P < 0.01$) when compared to the clean air, while the opposite was observed when they were exposed to healthy wheat plants ($\chi^2 = 4.52$, $P < 0.05$).

The proportion of individuals that were attracted by odours from healthy wheat plants mixed with aphid-infested pea plants was significantly higher when compared to the clean air ($\chi^2 = 4.49$, $P < 0.05$). No significant differences were observed between the other treatments and the clean air (Fig. 4.).

E. balteatus behavioural observations

The combination of WA, PW and WA induced high frequencies of searching by *E. balteatus* females

compared to the combination of WW, WW and WW (Student's t-test: $t = 2.29$, $P < 0.05$) (Fig. 5.).

There were significant difference in acceptance frequencies of *E. balteatus* females as follow groups: PA, PA, PA and WA, WA, WA (Student's t-test: $t = 2.42$, $P < 0.05$), WW, PW, WW and WW, WW, WW (Student's t-test: $t = 2.22$, $P < 0.05$), WA, PA, WA and WW, WW, WW (Student's t-test: $t = 2.43$, $P < 0.05$).

Table 1. The different model (combination) between wheat and pea.

Series	A			B		
	1	2	3	4	5	6
Test 1	PW	PW	PW	WW	WW	WW
Test 2	PW	PW	PW	WA	WA	WA
Test 3	PA	PA	PA	WW	WW	WW
Test 4	PA	PA	PA	WA	WA	WA
Test 5	WW	PW	WW	WW	WW	WW
Test 6	WW	PW	WW	WA	WA	WA
Test 7	WW	PA	WW	WW	WW	WW
Test 8	WW	PA	WW	WA	WA	WA
Test 9	WA	PW	WA	WW	WW	WW
Test 10	WA	PA	WA	WA	WA	WA
Test 11	WA	PA	WA	WW	WW	WW
Test 12	WA	PW	WA	WA	WA	WA

PW: one pot of pea without aphids, PA=one pot of pea infested with aphids(50 ints), WW=one pot of wheat without aphids, WA=one pot of wheat infested with aphids(50 ints) A and B represent zone A and B respectively, 1,2,3,4,5 and 6 represent the number of site in net-cage.

Moreover, the oviposition frequencies related to the pea plant infested by related aphid or not were higher than the ones observed with wheat plants (Fig. 5. Student's t-test: $t = 2.38$, $P < 0.05$).

Field experiments

Diversity and abundance of aphids

Among the recorded aphid species, *M. dirhodum* and *S. avenae* were predominant on wheat, while *A. pisum* was predominant on pea plants. The abundance of *A. pisum* was far higher than the one from cereal aphids in both visual observations and traps (Fig. 6. and Table 2). The population dynamics of *M. dirhodum*, *S. avenae* and *A. pisum* exhibited the same trends.

Population densities of *M. dirhodum*, *S. avenae* and *A. pisum* reached their peak in all treatments on June 15th, June 22nd and June 22nd, respectively.

According to visual observations, *M. dirhodum* was significantly more abundant in WM than in SWP and in WMP both on peak occurrence period and on the whole experimental duration (peak: $F_{2,6} = 37.90$, $P < 0.01$; total: $F_{2,6} = 20.44$, $P < 0.01$). Similarly, a significant difference for *M. dirhodum* in traps was also detected among treatments (peak: $F_{2,6} = 21.43$, $P < 0.01$; total: $F_{2,6} = 30.43$, $P < 0.01$). Consistently with the results of *M. dirhodum*, the abundance of *S. avenae* on wheat plants was significantly

higher in WM than in SWP and WMP both on peak occurrence period and on the whole experimental duration (peak: $F_{2,6} = 34.78$, $P < 0.01$; total: $F_{2,6} = 27.15$, $P < 0.01$). Similar results were found for *S. avenae* in yellow traps (peak: $F_{2,6} = 61.27$, $P < 0.01$; total: $F_{2,6} = 51.52$, $P < 0.01$).

In addition, according to both trapping and visual observations, population densities of *A. pisum* were significantly reduced by mixing and strip intercropping wheat with pea (Fig. 5.). The abundance of *A. pisum* was significantly lower in SWP and WMP than in PM (trap peak: $F_{2,6} = 32.22$, $P < 0.01$, total: $F_{2,6} = 38.00$, $P < 0.01$; observation peak: $F_{2,6} = 31.38$, $P < 0.01$; total: $F_{2,6} = 79.64$, $P < 0.01$).

Table 2. Diversity and abundance of aphids and related beneficials recorded in yellow traps in different crop systems.

Species	Treatments				
	Wheat-pea mixing	Wheat-pea strips	Wheat monoculture	Pea monoculture	% ^a
Aphids					
<i>Metopolophium dirhodum</i> (W.)	578	437	949	0	67.6
<i>Sitobion avenae</i> (F.)	89	43	276	0	14.0
<i>Acyrtosiphon pisum</i> H.	64	131	0	339	18.4
Total	731	611	1225	339	
Relative abundance (%)	25.1	21.0	42.2	11.7	
Ladybirds					
	10.83 ^b				
<i>Coccinella 7-punctata</i> L.	5	17	8	9	40.2
<i>Harmonia axyridis</i> (P.)	5	14	8	18	46.4
<i>Propylea 14-punctata</i> (L.)	0	2	0	0	2.1
<i>Harmonia 4-punctata</i> (P.)	2	0	0	0	2.1
<i>Calvia 14-guttata</i> (L.)	1	1	1	4	7.2
<i>Hippodamia variegata</i> (G.)	1	1	0	0	2.0
Total	14	35	17	31	
Hoverflies					
	43.08 ^b				
<i>Episyrphus balteatus</i> (D.)	88	112	69	56	84.2
<i>Scaeva pyrastris</i> (L.)	0	3	2	0	1.3
<i>Sphaerophoria scripta</i> (L.)	5	8	4	0	4.4
<i>Melanostoma scalare</i> (F.)	0	1	2	0	0.8
<i>Metasyrphus corolla</i> (F.)	8	15	4	9	9.3
Total	101	139	81	65	
Lacewing fly					
	46.09 ^b				
<i>Chrysoperla carnea</i> (S.)	115	142	74	82	100.0
Total predators	230	316	172	178	
Relative predator abundance (%)	25.6	35.3	19.2	19.9	

^aRelative occurrence of each species by family

^bRelative occurrence of each family in beneficial populations.

Diversity and abundance of aphid natural enemies

Lacewings were the main aphid natural enemies trapped (46.1%), followed by hoverflies (43.1%) and ladybirds (10.8%). *C. carnea*, *E. balteatus* and *H. axyridis* were the predominant recorded species (Table 2).

Lacewings reached their occurrence peak in all treatments on June 15th. Their abundance in each treatment was low before June 8th even if they were significantly more abundant in SWP than in others three treatments at that period ($F_{3,8} = 15.00$, $P < 0.05$).

Taking into account the whole experimental duration, lacewings were significantly more abundant in SWP and WMP when compared to both monocultures ($F_{3,8} = 8.73, P < 0.05$)

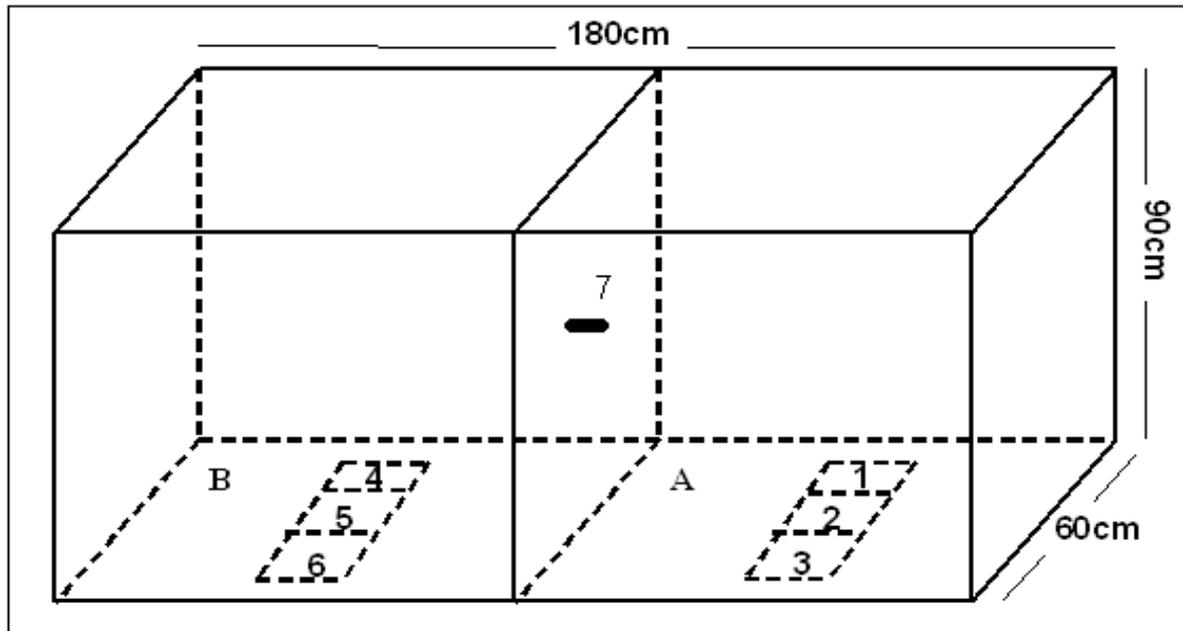


Fig. 1. Schematic net-cage used for *E. balteatus* behavioural assays in response to cues originating from wheat and pea. 1, 2, 3, 4, 5 and 6 represented sites for plant container setting, 7 represented site where *E. balteatus* were released, A: the combination of plants A, B: the combination of plants B.

The occurrence peak of hoverflies occurred from 22nd to 29th of June. There was no significant difference among treatments in population densities before this period. After that, their abundance was significantly higher in SWP and WMP when

compared to both monocultures ($F_{3,8} = 114.43, P < 0.05$). Taking into account the whole experimental duration, hoverflies were significantly more abundant in SWP followed by WMP, WM and PM ($F_{3,8} = 11.74, P < 0.05$).

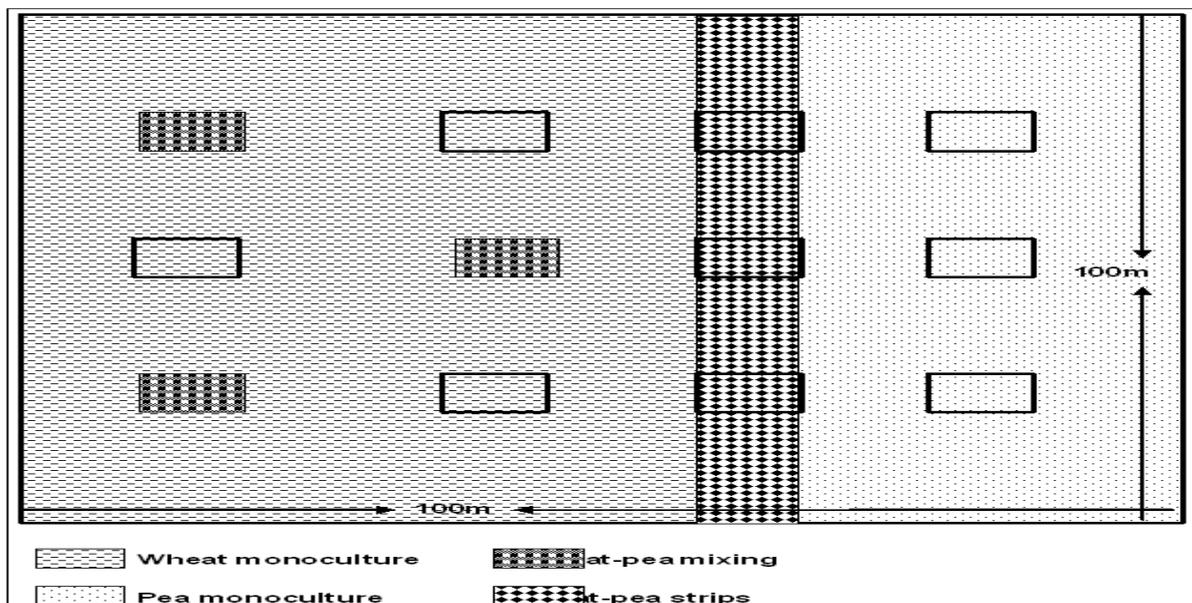


Fig. 2. Experimental field set-up to assess different kinds of wheat and pea associations on aphid and related beneficial abundance and diversity.

As for ladybirds, their abundance was significantly higher SWP and WMP when compared to both monocultures when taking into account the whole experimental duration, the ($F_{3,8} = 12.39, P < 0.05$).

Discussion

Behavioural assays

Semiochemical-mediated host selection has been shown to occur in several insect species (De Moraes *et al.*, 2001; Han and Chen, 2002; Sema Gencer *et al.*, 2009; Verheggen *et al.*, 2008).

In the case of aphids, volatile organic compounds (VOCs) are important olfactory cues for them to locate their host plants (Döring, 2014).

Our results show that winged *S. avenae* significantly prefer odours from healthy wheat or pea plants alone when compared to the ones from different wheat-pea mixtures combined with aphid infestations.

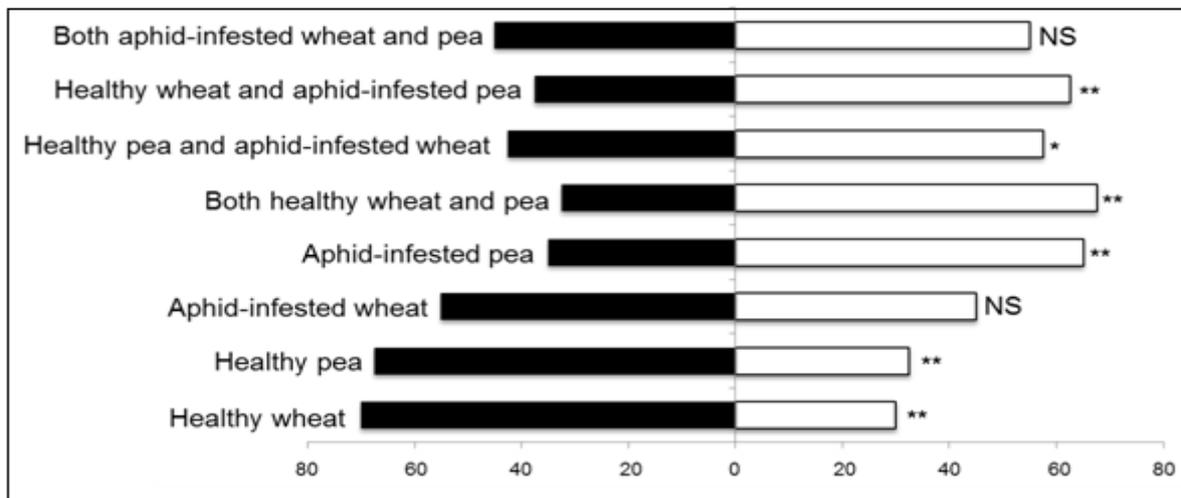


Fig. 3. Odour source selection (relative proportion of individuals choosing each arm) by winged *Sitobion avenae* to different healthy/aphid-infested wheat and/or pea associations. ** $P < 0.01$, * $P < 0.05$, NS: not significant.

Similarly, it has been reported that odours from uninfested maize seedlings were significantly more attractive to the leafhopper, *Cicadulina storeyi* China than odours from *C. storeyi*-infested seedlings (Oluwafemi *et al.*, 2011). When tested individually for behavioural activity, VOCs from *C. storeyi*-infested seedlings such as methyl salicylate, (E)-caryophyllene, and (E)- β -farnesene were repellent for *C. storeyi*. Other behavioural assays also revealed that several VOCs are released from herbivore-induced tobacco plants exclusively at night and are highly repellent to female moths from the species *Heliothis virescens* (Fabricius) (De Moraes *et al.*, 2001). In our assays, odours from the mixture of healthy wheat and pea were also deterrent to *S. avenae*. Similarly, Xie *et al.* (2012) showed that *S. avenae* prefer wheat plant odours alone than blended odours of wheat intercropped with mung bean.

Herbivore-induced plant volatiles are also important foraging cues for predators (Dicke *et al.*, 1990). *E. balteatus* foraging and reproductive behaviours are known to be enhanced by volatiles emitted from aphid-infested plants (Harmel *et al.*, 2007). For example, (Z)-3-hexenol and (E)- β -farnesene can induce higher frequencies of *E. balteatus* female searching and acceptance behaviour (Alhmedi *et al.*, 2010; Almohamad *et al.*, 2008). Similar results were obtained in our study as volatiles from wheat-pea mixtures combined with aphid infestations were more attractive for *E. balteatus* females, increasing their frequencies of acceptance and oviposition. Surprisingly, no significant attraction was found for *H. axyridis* females, excepting when healthy wheat was combined with aphid-infested pea.

For example, olfactometer experiments showed that adult *C. septempunctata* were significantly more attracted by odours from barley mixed with the common weeds *Cirsium arvense* (Linnaeus) Scop. and *Elytrigia repens* (Linnaeus) Nevski. than barley alone (Ninkovic and Pettersson, 2003; Pettersson *et al.*, 2005). Similarly, Glinwood *et al.* (2009) reported that *C. septempunctata* were more attracted to combined odours from certain barley cultivars than

each cultivar alone. In another study, *C. septempunctata* responded positively to volatiles from aphid-infested barley plants and from previously aphid-infested plants but not to volatiles from uninfested plants (Ninkovic *et al.*, 2001). Despite our results, this suggests that olfactory cues from diversified plant stands can be important mechanisms in predator attraction to sites with a complex botanical diversity.

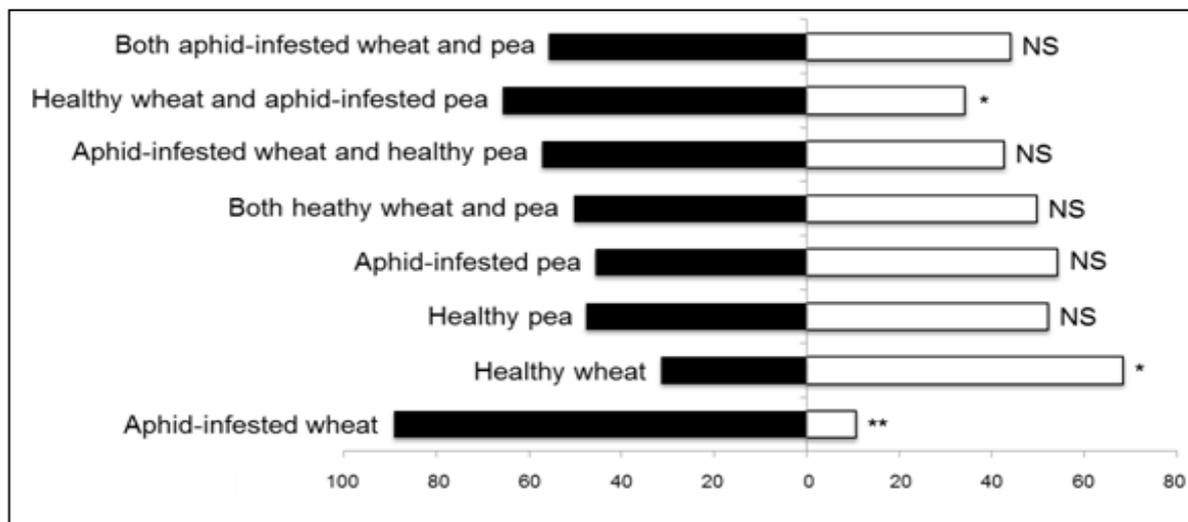


Fig. 4. Odour source selection (relative proportion of individuals spending more time in the olfactometer arms) by *Harmonia axyridis* females in response to different healthy/aphid-infested wheat and/or pea associations in dual-choice experiments. ** $P < 0.01$, * $P < 0.05$, NS: not significant.

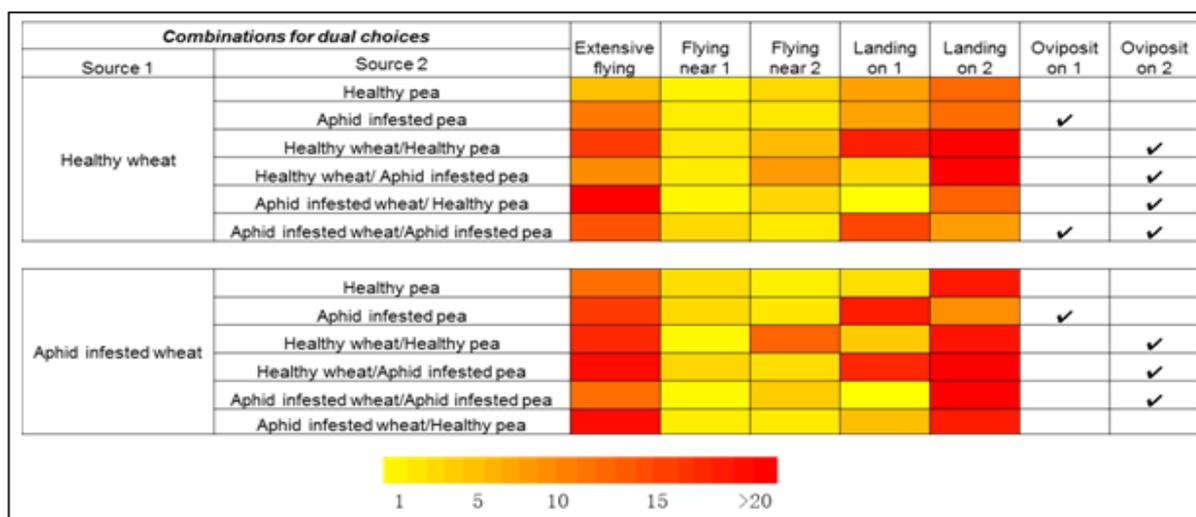


Fig. 5. Heatmap to illustrate behavioural changes (flying, landing and ovipositing on plants) of *Episyrphus balteatus* females in relation to different dual choice experiments including healthy or aphid-infested wheat and/or pea. Yellow to red colors correspond to increasing mean durations for hoverfly related activities.

Field trial

The aim of field habitat management is to create suitable ecological infrastructures within the agricultural landscape to decrease pest pressure on crops and provide resources for natural enemies such as alternative prey or hosts and shelter (Landis *et al.*, 2000). According to Root's *natural enemies* hypothesis, generalist and specialist natural enemies are expected to be more abundant in polycultures and therefore suppress herbivore population densities more in polycultures than in monocultures (Root, 1973). In our study, the abundance of lacewings, hoverflies and ladybirds was

improved when pea was associated to wheat. This could partly explain why the populations of cereal and pea aphids were both decreased significantly when compared to monocultures. Other factors such as the physical obstruction (Perrin and Phillips, 1978) and visual camouflage (Smith, 1969, 1976) of host plants may have contributed to reduce the abundance of aphids in wheat-pea associations. Other similar studies showed that growing pea between rows of wheat can reduce the populations of *S. avenae* and enhanced those from natural enemies (Zhou *et al.*, 2009a; Zhou *et al.*, 2009b).

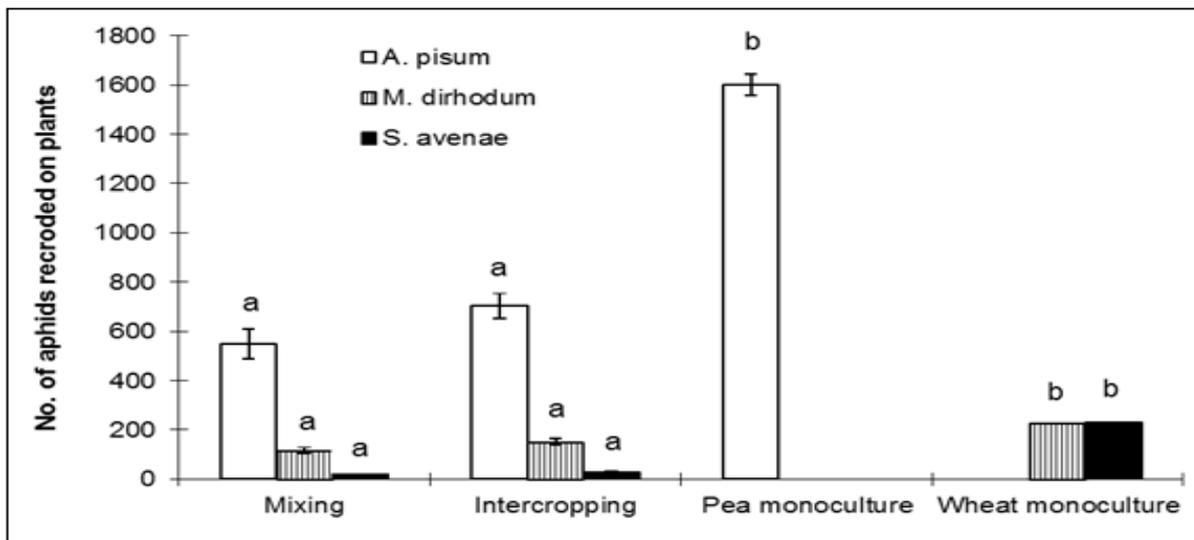


Fig. 6. Abundance of aphids (Mean±SEM) recorded by visual observation in the different treatments during the whole sampling period. Different letters indicate significant differences at $P < 0.05$.

Overall, the above findings suggest that intercropping plant species as an habitat management strategy can be interesting to reduce aphid populations and increase aphidophagous beneficials. The combination of wheat and pea, with or without related aphid species, improved the frequencies of acceptance and oviposition by *E. balteatus* females, and also reduced the attraction of *S. avenae*. Further behavioural studies could focus on other important aphidophagous such as lacewings and aphid parasitoids. Results from our behavioural assays were consistent with those from the field trial, supporting the idea that wheat-pea associations are an efficient tool for aphid biological control.

Therefore, it could be seen as an alternative method to reduce the reliance on insecticides in agroecosystems.

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