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## Keywords

Conservation biological control, *Cucumis melo*, ecological engineering, field margins, habitat manipulation, natural enemies

## Abstract

Flower strips appear to be a promising lever for promoting pest control but a careful selection of the plant species used is needed prior to implementation to avoid possible negative side effects. In the case of open field melon crops, the main pitfall would be to generate aphid and aphid-borne virus reservoirs near the crops. Combining biotests under controlled conditions and data from the literature, we assessed 18 candidate plant species, and ruled-out those posing a potential risk of hosting *Aphis gossypii* (melon pest and virus vector), *Myzus persicae* (virus vector) and/or viruses (*Cucurbit aphid-borne yellows virus* (CABYV), *Cucumber mosaic virus* (CMV), *Watermelon mosaic virus* (WMV) and *Zucchini yellow mosaic virus* (ZYMV)). Five plant species made it through the selection process: cornflower, grass pea, sainfoin, salad burnet and sweet marjoram. Flower strips sown with a mix of these five plant species were evaluated in a five-year field experiment. They displayed a flowering continuum likely to provide a food resource to natural enemies throughout the growing season. Their potential to host natural enemies was compared to those of grass strips and bare soil by monitoring generalist and specialist predators within the different field margins and melon crop. Flower margins supported significantly more of these natural enemies than grass margins and bare soil. All predator taxa analyzed responded positively to the floral resources displayed. Spiders were 3.2 times more abundant in pitfall traps placed in flower margins than in bare soil. Generalist predators and aphid specialist predators collected using a vacuum sampler were 5.5 and 9.1 times more abundant in flower margins than in bare soil, respectively. Interception traps set for weekly periods showed that coccinellid and syrphid fluxes were significantly enhanced near flower margins.

## 1. Introduction

Aphids and aphid-borne viruses can cause severe economic damage to open field vegetable crops. In conventional cropping systems, synthetic insecticides are often the first option for controlling aphids. However their efficiency is frequently challenged by the development of resistant clones (Bass et al., 2015) and their usage is increasingly questioned due to environmental and health risks. In addition, they are of little relevance to limit non-persistently transmitted viruses, where acquisition and inoculation occur in a matter of seconds (Perring et al., 1999). Cultural practices such as the use of plastic mulches or row covers may limit both aphid colonization and the transmission of aphid-borne viruses but only provide temporary protection and lead to plastic waste (Lecoq and Desbiez, 2012). Whenever available, resistant cultivars are the easiest, most efficient and environmentally friendly way to manage pests and diseases, but genetic resistance durability can be jeopardized by the emergence of adapted clones. A complementary way of reducing aphid and virus damage within an open field crop can be to take advantage of ecological services provided by the environment. For example, Losey and Vaughan (2006) estimated that the annual value of pest control by wild native insects is $4.5 billion in the United States. Unfortunately, agricultural landscapes are rarely optimal environments for natural enemies. The excessive use of insecticides and the lack of alternative food resources generally limit the performance of natural enemies (Tscharntke et al., 2016). Thus increasing attention is being paid to conservation practices that enhance the survival, fecundity, longevity and behavior of natural enemies. In particular, habitat management seeks to alter the habitats within or around crops to improve the availability of the resources required for optimal performance by natural enemies (Gurr et al., 2017; Landis et al., 2000). To achieve pest control it is crucial to provide nectar, pollen, alternative hosts or prey, shelter, to parasitoid

and predator insects. Although flowering plants provide many of these food resources, habitat manipulation should be more than "chocolate-box ecology" (Gurr et al., 2004). Floral displays of plants picked at random can be at best ineffective, at worst detrimental by favoring pest populations over beneficial organisms. Thus a rigorous evaluation of the candidate insectary plants prior to flower strip implementation is needed to avoid these adverse effects.

Melon, particularly the Charentais-type (*Cucumis melo* var. *cantalupensis*), is a very popular fruit in France. It is cultivated between March (early tunnel plantings) and September (late open field plantings) in three main production areas: South-East, South-West and Central-West. Nearly 40% of the national production is in the South-East (286 000 t, 14000 ha in 2017, www.agreste.agriculture.gouv.fr). Open field melon crops are regularly impacted by biotic stresses, among which aphids and aphid-borne viruses. The melon aphid *Aphis gossypii* Glover (Hemiptera: Aphididae) is the only aphid species colonizing melon crops in France, causing leaf-curling, stunting and even plant death when colonization is intense. *Myzus persicae* Sulzer (Hemiptera: Aphididae) has not yet been found to colonize melon crops in France, but it is the most important aphid virus vector, able to transmit over 100 plant viruses (Blackman and Eastop 2000). Both aphid species are efficient vectors for four viruses frequently observed on melon crops in France: *Cucurbit aphid-borne yellows virus* (CABYV, Polerovirus, Luteoviridae), *Cucumber mosaic virus* (CMV, Cucumovirus, Bromoviridae), *Watermelon mosaic virus* (WMV, Potyvirus, Potyviridae) and *Zucchini yellow mosaic virus* (ZYMV, Potyvirus, Potyviridae). WMV and ZYMV are the most harmful viruses, causing mosaic symptoms on leaves, plant stunting and reduced fruit yield, but also, when infection is severe, leaf deformation such as filimorphism, mosaic on fruits, coupled with marbling and hardening of the flesh for ZYMV (Lecoq and Desbiez, 2012).

In France as elsewhere in Europe, increasing importance is being granted to habitat management, not only in organic farming systems, but also in conventional systems due to the

progressive reduction of pesticide usage imposed by changes in regulation. For melon producers, flower strips could be a promising lever for controlling aphids. Several seed companies sell blends which are supposed to boost biological control services. These advised blends are identical regardless of crop and are not specific for melon crops. Yet pathogen corteges differ a lot depending on the crops, so botanical composition should be crop-specific to avoid negative side effects. In the case of open field melon crops, the main pitfall would be to generate aphid and virus reservoirs near the crop by sowing plant species likely to host melon aphids and viruses.

In this study, we focused on the selection and evaluation of flowering species appropriate for melon crops. The first step was to screen a set of candidate plants under controlled conditions to design a mix that minimizes the risk of hosting aphids (*A. gossypii* and *M. persicae*) and viruses (CABYV, CMV, WMV and ZYMV). The second step was to evaluate, under field conditions, the potential of the corresponding flower strips to enhance aphid predator abundance within field margins and melon crop.

## 2. Materials and methods

### 2.1. Plant screening under controlled conditions

#### 2.1.1. Plant species shortlist and plant rearing

Twenty plant species from 13 families were selected for the experiments (Table S1). Melon (*Cucumis melo* L.) and pepper (*Capsicum annuum* L.) were used as controls.

Virus-free plants were grown in an insect-proof greenhouse without pesticides. Seedlings were prepared in flats containing a peat/coco coir substrate (080 Klasmann-Deilmann France, Bourgoin Jallieu, France) and irrigated daily with bore water. Plantlets were transplanted individually to plastic pots (9 cm wide × 8 cm high) containing a peat/clay substrate (404 Klasmann-Deilmann France) and irrigated daily with bore water, the pH of which was adjusted to 5.8. Plants were used at the vegetative stage (3-8 week old depending on plant species, Table S1). During biotests, plants were irrigated daily with a nutrient solution (Soluveg Essentiel 16-5-25, Angibaud & Spécialités, La Rochelle, France).

#### 2.1.2. Aphid rearing

Aphids were reared on virus-free plants in growth cabinets equipped with LED tube lights (4000K) under a photoperiod of 16:8 (L:D) h. *Aphis gossypii* (clone NM1, Thomas et al., 2012) was reared on 3-5 week old melon plants (cv. Védrantais) at 24/21±1°C day/night. *Myzus persicae* (clone Patho) was reared on 5-8 week old pepper plants (cv. Yolo Wonder) at 21±1°C day/night. Four weeks before experiments, mass rearing was shifted to synchronous rearing in order to manipulate 7 day-old apterous female cohorts.

2.1.3. Plant-aphid interaction biotests

Each plant species was evaluated for its capacity to host two aphid species: *A. gossypii* and *M. persicae*. For each aphid species, no-choice settling tests were carried out on 10 plants per plant species, each plant tested constituted a replicate. Ten 7 day-old apterous females were deposited per plant. After 24h, settled females were counted and removed. The acceptance rate was calculated as the number of recovered females divided by 10. After six additional days, the offspring produced during the initial 24h period was counted. The reproductive rate was calculated as the number of offspring divided by the number of recovered females. Together, the acceptance rate and the reproductive rate were used to assess the capacity of the tested aphid species to accept the tested plant species as a suitable host. Results were compared to those obtained on plant species used for aphid rearing, which were considered as reference plant species (melon for *A. gossypii*, pepper for *M. persicae*).

2.1.4. Plant-virus interaction biotests

Each plant species was evaluated for its capacity to host the most frequently observed viruses in melon crops in France: CABYV, CMV, WMV and ZYMV. For CABYV and CMV, one isolate was used; for WMV and ZYMV, two isolates reflecting the recent changes in the genetic structure of these viruses in France were used (Table 2). For each viral isolate, biotests were conducted on eight plants per plant species, each plant tested constituted a replicate. Melon (cv. Védrantais) was used as positive control.

The persistently aphid-transmitted CABYV was tested through aphid transmission using *A. gossypii* as vector. Virus sources were infected melon plants (cv. Védrantais). Virus-free aphids reared as in Section 2.1.2. were collected with a fine-tip paint brush and transferred onto virus sources. After a 48h acquisition access period (AAP), groups of 10 aphids were gently transferred to virus-free test plants for a 48h inoculation access period (IAP). At the end of the

153 IAP, aphids were killed by spraying the plants with two systemic insecticides (0.5 ml/l NUPRID
154 200, Nufarm SAS) at 24h intervals. Plants were then placed in a dedicated greenhouse for 4-5
155 weeks of incubation.
156 The non-persistently aphid-transmitted CMV, WMV and ZYMV were tested through
157 mechanical inoculation. Virus sources were infected zucchini plants (*Cucurbita pepo* L. cv.
158 Diamant). Inoculum was prepared by grinding 2 g of young leaf tissue with a mortar and pestle
159 in 8 ml of a solution containing 0.03M $Na_2HPO_4$ with 0.2% DIECA. Carborundum (75 mg/ml)
160 and activated charcoal (75 mg/ml) were added before rub-inoculation of test plants. Plants were
161 rinsed with tap water and placed in a dedicated greenhouse for 3-5 weeks of incubation. Viruses
162 were detected using a double antibody sandwich enzyme-linked immunosorbent assay (DAS-
163 ELISA) with specific polyclonal antisera produced in our laboratory. Plants were considered
164 infected when the absorbance at 405 nm (Multiskan EX, Thermo Electron Corporation) was
165 three times above the mean value of the healthy controls (Schoeny et al., 2017). The
166 transmission rate was calculated as the number of infected plants to the total number of tested
167 plants.
168
169 2.2. *In situ* evaluation of field margins
170
171 2.2.1. Experimental design
172 Five field trials were conducted between 2011 and 2015 at the INRA St Paul experimental
173 station in Avignon (southeastern France, 43°54’53N, 4°52’59E) on a 1.3 ha plot edged north
174 and south by 6 m-high cypress trees. The experimental design consisted of a melon crop with
175 three modalities of field margin management: bare soil, flower strip and grass strip (Figure S1).
176 Each modality consisted of two strips 25.5 m apart: one ‘north strip’ and one ‘south strip’. Strips
177 were 55 m long and 3 m wide. The spatial organization of the modalities was defined randomly

each year. Flower strips were sown with a mix of five plant species selected after the above mentioned biotests: sainfoin (*Onobrychis viciifolia*, 22 kg/ha, 40%) grass pea (*Lathyrus sativus*, 16.5 kg/ha, 30%), salad burnet (*Sanguisorba minor*, 11 kg/ha, 20%), cornflower (*Centaurea cyanus*, 2.75 kg/ha, 5%) and sweet marjoram (*Origanum majorana*, 2.75 kg/ha, 5%). The proportions of each plant species in the mix were defined with the expertise of a seed seller. Grass strips were sown with ryegrass (*Lolium perenne*, 50 kg/ha). Flower and grass strips were sown during the second half of March (18-30 March) approximately two months before melon planting (24-31 May). Special attention was given to soil preparation and weed control to foster the establishment and growth of the field margins. Two tillage operations (disc harrowing before or after ploughing) were implemented in late autumn/winter. Just before sowing, a rotary harrow completed soil preparation to create a perfect seedbed. Flower and grass strips were sown with a portable spreader (421-S, Solo®). For the flower strips, to optimize seed germination and considering the difference in seed size of the five plant species selected, large seeds (sainfoin, grass pea, salad burnet) were sown in a first passage, followed by a comb harrow, the small seeds (cornflower, sweet marjoram) were sown in a second passage, completed by a roller. Flower and grass strips were irrigated with sprinklers (up to 63 mm per week depending on weather conditions) and hand-weeded when needed. The bare soil modality was maintained using mechanical weeding.

Melon crops were set up between the north and south strips after soil preparation with a rotary harrow. Charentais-type melon seedlings at the 1-3 leaf stage were planted in 16 rows (1.5 m row spacing) parallel to the strips on dark brown plastic mulch with drip irrigation (Figure S1). Basal PK fertilization (0-25-25) was applied during winter/early spring (250-300 kg/ha), complemented with ammonium nitrate (33-0-0) during spring in 2011 and 2012 (130 kg/ha). A monoammonium phosphate (12-61-0) fertigation was applied just after planting (100 kg/ha) to boost melon growth. No insecticides were applied during trials.

203

204 2.2.2. Plant development monitoring

205 The development of sown strips was monitored 2-3 times before melon planting with the
206 quadrat technique. A 1-$m^2$ wooden frame was randomly placed in the north and south strips and
207 the percentage of plant cover was visually estimated.

208 The flowering of flower strips was monitored 4-6 times after melon planting. Ten randomly
209 chosen plants per species were examined in one quadrat per strip in 2011 and 2012, in five
210 quadrats per strip in 2013, 2014 and 2015. Plants were considered at flowering stage when
211 displaying at least one open flower. Specific flowering rates were calculated as the number of
212 flowering plants divided by the number of examined plants.

213

214 2.2.3. Arthropod monitoring

215 To assess the biocontrol potential of the different field margins, we surveyed both generalist
216 (able to feed on various prey) and specialist (mainly aphidophagous) predators. Three
217 complementary trapping methods were used: pitfall traps to assess epigeal spiders moving on
218 the soil surface, vacuum sampling to assess arthropods active in the vegetation, and interception
219 traps to assess arthropods flying or moving from the margins to the crop (Figure S1).

220 Pitfall trapping was used at two dates in 2013 (27/06, 19/07) and 2014 (23/06, 23/07) when the
221 vegetation was fully developed. Traps were placed within each field margin strip (north and
222 south) and within the melon crop at two different positions (inter-rows 3 and 15) and left in
223 place for one week of monitoring. The experimental design comprised two and three replicates
224 per strip or position in 2013 and 2014, respectively. The data of 120 traps were analyzed.

225 Vacuum sampling was used at two dates in 2012 (21/06, 03/07), 2013 (20/06, 01/07) and 2014
226 (20/06, 10/07). At each date and for each modality, four samplings were made in the morning
227 in the melon crop (inter-rows 6 and 12) and field margins (for flower and grass strips: two in

north strips and two in south strips; for bare soil: four in the spontaneous vegetation of the pathway between the north cypress hedge and the trial, to avoid sampling dusty dry soil). For each sample, the pipe of the vacuum device (441, Solo®) equipped with a collection bag was placed 5 x 1s within the canopy.

Interception trapping was used in 2014 (13/06-18/07) and 2015 (29/05-10/07). Lightweight tent traps (BT2003, Bugdorm, MegaView Science Co., Taiwan) made of black polyester fabric (96x26 mesh/square inch, mesh aperture: 680 µm) were used (Figure 1a). They are 60 cm wide, 60 cm long and 60 cm high, with a dome-shaped window (45 cm wide and 27 cm high) in one panel. Insects entering the trap tend to fly upwards until they fall into the collecting bottle (500 ml) half-filled with a 30% ethanol solution with 5 µl/l detergent (Teepol 610 S, ref 86350, Sigma-Aldrich) to kill and preserve the catch. Four tent traps were set up (two facing the bare soil and two facing the flower strips) for 5-6 weeks of monitoring. Collecting bottles were changed weekly.

Arthropods collected by the three trapping methods were stored in 70% ethanol until taxonomic identification under a stereomicroscope. Generalist predators (Aeolothripidae (Thysanoptera), Anthocoridae, Lygaeoidae, Miridae and Nabidae (Hemiptera), Carabidae and Staphylinidae (Coleoptera), Dermaptera, Arachnids (spiders)) and specialist predators (Coccinellidae (Coleoptera), Neuroptera, Syrphidae (Diptera)) were identified to the taxonomic level required to know their feeding behavior and counted.

## 2.3. Data analysis

### 2.3.1. Biotests under controlled conditions

No-choice settling tests were performed to evaluate the capacity of 18 candidate plant species to host two aphid species (*Aphis gossypii* and *Myzus persicae*). For each aphid species, a Chi-

253 square test was conducted to determine if the plant species affected the acceptance rate
254 (proportion of aphids recovered 24h after deposition). When the null hypothesis of equality was
255 rejected, the Marascuilo procedure for pairwise multiple comparisons was applied. The effect
256 of the plant species on the reproductive rate (number of offspring per recovered aphid) was
257 investigated through nonparametric Kruskal-Wallis tests. When the null hypothesis of equality
258 was rejected, Dunn's pairwise multiple comparisons were performed using the Bonferroni
259 correction.
260 Transmission tests were performed to evaluate the capacity of the candidate plant species to
261 host four viruses (CABYV, CMV, WMV and ZYMV). For each virus, a Chi-square test was
262 conducted to determine if the plant species affected the transmission rate. When the null
263 hypothesis of equality was rejected, the Marascuilo procedure for pairwise multiple
264 comparisons was applied.
265 All statistical analyses were performed using XLSTAT (version 2015.4.01, Addinsoft, Paris).
266
267 2.3.2. Field experiments
268 Depending on the trapping method used to monitor aphid predators and thus the main taxa
269 caught, statistical analyses were conducted on different categories: spiders for pitfall traps;
270 generalist predators and specialist predators for vacuum sampling and flight interception traps.
271 For this latter trapping method, the abundance of specialist predators was further separated into
272 Coccinellidae, Neuroptera and Syrphidae.
273 For pitfall trapping and vacuum sampling, we first tested the independence of catches between
274 dates and years using a Spearman's correlation test (for each kind of field margin separately).
275 As no significant correlation was found, we considered each strip (north and south) and each
276 sampling date as independent values and computed mean values accordingly. We then tested
277 the effect of the management type (flower strip/grass strip and bare soil) and sampling zone

(margin/crop) on abundances using two-way ANOVA (no heteroscedasticity was detected) followed by post hoc multiple comparisons (Tukey HSD). For interception trapping, the effect of the management type (flower strip/bare soil) was investigated through a nonparametric Wilcoxon signed-rank test performed between paired values (n=22). All computations were carried out using R software.

## 3. Results

### 3.1. Plant screening under controlled conditions

#### 3.1.1. Evaluation of the risk of aphid infestation

For *A. gossypii*, the mean percentage of aphids recovered after 24h (acceptance rate) ranged from 1 to 95% and the mean number of offspring per recovered aphid (reproductive rate) ranged from 0 to 18.4 (Table 1). The acceptance rates obtained for basil, bullwort and French marigold were not significantly different from that obtained for melon ($\chi^2$ = 1729, df = 18, p-value < 0.0001). The reproductive rates for borage, bullwort, lacy phacelia were not significantly different from that obtained for melon (Kruskal-Wallis test: K = 293, df = 18, p-value < 0.0001). For *M. persicae*, the mean acceptance rate ranged from 0 to 97% and the mean reproductive rate ranged from 0 to 6.0 (Table 1). The acceptance rates obtained for borage, bullwort, corn marigold, dill, field marigold were not significantly different from that obtained for pepper ($\chi^2$ = 1832, df = 19, p-value < 0.0001). The reproductive rates for borage, corn marigold, dill, field marigold, lacy phacelia were not significantly different from that obtained for pepper (K = 293, df = 19, p-value < 0.0001). A plant species was considered as a suitable host for an aphid species when its acceptance rate and/or reproductive rate were not significantly different from the reference rates obtained on plant species used for aphid rearing (melon for *A. gossypii* and pepper for *M. persicae*). Thus, in the case of *A. gossypii*, five species were accepted as hosts: basil, borage, bullwort, French marigold and lacy phacelia (Table 1). *M. persicae* was able to use six species as hosts: borage, bullwort, corn marigold, dill, field marigold and lacy phacelia (Table 1). Considering the potential impact they could have on *A. gossypii* and/or *M. persicae* populations and subsequently on virus transmission, eight plant species were considered high

risk for melon crops: basil, borage, bullwort, corn marigold, dill, field marigold, French marigold and lacy phacelia.

3.1.2. Evaluation of the risk of virus infection

A plant species was considered a virus host when its transmission rate was not significantly different to the reference rate obtained on melon (Table 2). Dill was the only host for CABYV ($\chi^2$ = 58, df = 18, p-value < 0.0001). Eleven plant species were infected with CMV: basil, borage, buckwheat, corn marigold, cornflower, French marigold, lacy phacelia, marigold, nigella, sweet marjoram and white campion ($\chi^2$ = 139, df = 18, p-value < 0.0001). Six plant species were infected with both WMV isolates: buckwheat, bullwort, dill, field marigold, lacy phacelia and nigella (LL1A: $\chi^2$ = 152, df = 18, p-value < 0.0001; LL2B3: $\chi^2$ = 153, df = 18, p-value < 0.0001). Three plant species were infected with both ZYMV isolates: bullwort, dill and nigella (E9 and 124L11: $\chi^2$ = 155, df = 18, p-value < 0.0001). Taking into account the relative harmfulness of the four viruses, plant species capable of hosting WMV and/or ZYMV were considered a high risk for melon crops. These were buckwheat, bullwort, dill, field marigold, lacy phacelia and nigella.

3.1.3. Combining risks of aphid infestation and virus infection to select plant species

Ten plant species, which showed a high risk for both aphid infestation and virus infection according to biotests, were discarded from the selection: basil, borage, buckwheat, bullwort, corn marigold, dill, field marigold, French marigold, lacy phacelia and nigella. Three additional plant species previously reported to be aphid hosts (Table S1) were also discarded: marigold, ryegrass and white campion.

330 The remaining five species (cornflower, grass pea, sainfoin, salad burnet and sweet marjoram)
331 were selected and combined in a seed mix with the expertise of a seed seller and tested *in situ*.
332
333 3.2. *In situ* evaluation of the flowering capacity of the seed mix
334
335 Over the course of our five-year experiment, cornflower, grass pea, sainfoin, salad burnet
336 established first and constituted the higher vegetation strata; sweet marjoram generally
337 appeared later, constituting the low stratum. At melon planting, nearly all strips had 100% plant
338 cover. Flowering spanned the entire melon cropping period (Table 3). Grass pea was generally
339 the only flowering plant species at melon planting. It started to flower around mid-May and
340 was abundant until the end of June. The flowering of cornflower generally started shortly after
341 melon planting and lasted until the end of July with a peak at mid-crop. Sainfoin and salad
342 burnet flowered between mid-June and the end of July. Sweet majoram had a short flowering
343 period centered around mid-July. Thus, the five-species mix allowed a flowering continuum
344 likely to provide a food resource continuum conducive to the development of natural enemies.
345
346 3.3. *In situ* evaluation of aphid predator abundance in field margins and melon crop
347
348 3.3.1. Pitfall trapping
349 Both the management type and the sampling location (field margin vs crop) had a significant
350 effect on spider abundance (p-value = 0.0018 and p-value = 0.0020, respectively). Within the
351 field margins, the mean number (± SEM) of spiders trapped per week was significantly higher
352 in the flower strip (36.1 ± 2.7) than in bare soil (11.3 ± 2.3) which corresponds to an average
353 increase of 219% (Figure 2). Spider abundance was intermediate in grass strip (22.6 ± 1.6)

(Figure 2). Within the melon crop, no difference was observed between the field margin management modalities. The mean numbers of spiders trapped per week ranged from 10.0 ± 1.0 to 15.7 ± 4.0, i.e. similar to that assessed in bare soil (Figure 2).

3.3.2. Vacuum sampling

The abundance of generalist predators was significantly affected by both the management type (p-value = 0.026) and the sampling location (p-value < 0.001). There was also a significant management type*sampling location interaction (p-value = 0.037) indicating that the effect of management type changed according to sampling location. Within the field margins, the mean number of generalist predators trapped per suction was significantly higher in the flower strip (18.3 ± 5.4) than in bare soil (3.3 ± 0.6) which corresponds to an average increase of 455% (Figure 3a). The abundance of generalist predators was intermediate in grass strip (13.1 ± 4.1) (Figure 3a). Within the melon crop, in contrast, the effect of management type was not significant with abundances ranging from 3.2 ± 0.7 to 4.1 ± 0.8 individuals trapped per suction (Figure 3a).

The abundance of specialist predators was significantly affected by both the management type (p-value < 0.001) and the sampling location (p-value = 0.0045). There was also a significant management type*sampling location interaction (p-value = 0.035). Within the field margins, the mean number of specialist predators trapped per suction was significantly higher in the flower strip (5.8 ± 1.0) than in the grass strip (2.0 ± 1.1) or bare soil (0.6 ± 0.3) which corresponds to an average increase of 813% in flower strip compared to bare soil (Figure 3b). Within the melon crop, the effect of management type was not significant with abundances ranging from 0.5 ± 0.2 to 2.0 ± 0.8 individuals trapped per suction (Figure 3b). Specialist

predators collected by vacuum sampling were mostly coccinellids (91%), rarely lacewings (8%) and syrphids (1%) (data not shown).

3.3.3. Interception trapping

Tent traps facing flower strips intercepted significantly more aphid predators than those facing bare soil: +95% for generalist predators (Wilcoxon signed-rank test: V = 191, p-value = 0.009), +174% for specialist predators (V = 166, p-value = 0.005) (Figure 4). Specialist predators (coccinellids, lacewings, syrphids) represented 20-27% of total predators. The impact of the field margin management type varied depending on the taxa: the mean numbers of coccinellids and syrphids trapped per week were significantly enhanced near flower strips compared to bare soil (V = 162, p-value = 0.034 and V = 133.5, p-value = 0.001, respectively), whereas the effect on lacewings was not significant (V = 42, p-value = 0.15).

## 4. Discussion

In this study, we implemented a multi-step plant selection process to design flower strips adapted to melon crops. In a first step, we evaluated the capacity of 18 candidate plant species to host *A. gossypii* and/or *M. persicae*. Host plants could harbor and increase the populations of these aphid species, thus generating pest and vector reservoirs near the crop. Based on our results under controlled conditions, eight plant species were considered a high risk for aphid infestation for melon crops and ruled out: borage, bullwort, corn marigold, dill, field marigold, French marigold and lacy phacelia. These are new host descriptions for *A. gossypii* (bullwort, lacy phacelia) and *M. persicae* (borage) compared to previous reports (Blackman and Eastop, 2006; Kavallieratos et al., 2007). In a second step, we evaluated the capacity of the same 18

candidate plant species to host the most frequently observed viruses in melon crops in France: CABYV, CMV, WMV and ZYMV. Under controlled conditions, 15 plant species showed systemic infection by at least one virus. These are new host descriptions for CABYV (dill), CMV (nigella, sweet marjoram), WMV (buckwheat, dill, field marigold and nigella) and ZYMV (dill, nigella) compared to the literature (Desbiez and Lecoq, 1997; Edwardson and Christie 1991; Edwardson and Christie, 1997; Lecoq et al., 1992). For CMV, this extends the already tremendous host list reporting 1287 species in 518 genera of 100 families (Edwardson and Christie, 1997). Taking into account the relative harmfulness of the four viruses (Lecoq and Desbiez, 2012), plant species capable of hosting WMV and/or ZYMV were ruled-out as a priority: buckwheat, bullwort, dill, field marigold, lacy phacelia and nigella. Morales et al. (2006) adopted a similar precautionary principle to select insectary plants suitable for growing in tomato crops and discarded plant species likely to be a reservoir for Potato virus Y and CMV. In a third step, we combined biotest results and literature to take into account potential aphid and virus infection risks not revealed in our study.

Like in a "guess who?" game, plant species presenting a risk regarding *A. gossypii* (melon pest and virus vector) and *M. persicae* (virus vector), and a risk regarding the most harmful viruses (WMV and ZYMV) were in turn removed from the candidate list. The remaining five species (cornflower, grass pea, sainfoin, salad burnet and sweet marjoram) were selected and combined in a seed mix to be further tested *in situ*. It is noteworthy that outsiders made it through the selection while plant species frequently cited in habitat management literature were ruled-out during the process. Such was the case for buckwheat and lacy phacelia which have been given much attention since the 1990s due to the large quantities of nectar and pollen they produce (Fiedler et al., 2008; Laubertie et al., 2012; Robinson et al., 2008; White et al., 1995). The same applies for bullwort and dill, plants of the Apiaceae family considered some of the best nectar sources. Indeed, due to their small open flowers accessible to short-tongued insects, they attract

426 a guild of beneficial insects, including syrphids, small parasitic wasps but also lacewings and
427 coccinellids (Fiedler et al., 2008; Laubertie et al., 2012). These plant species, however, appeared
428 clearly unsuitable for melon crops. In addition to being a host for *A. gossypii* and *M. persicae*,
429 buckwheat and lacy phacelia combined risks to CMV and WMV, bullwort combined risks to
430 CMV, WMV and ZYMV, and dill exhibited the maximal virus risk, as a suitable host for
431 CABYV, CMV, WMV and ZYMV.
432 Under our pedoclimatic conditions, the five-species mix sown two months before melon
433 planting was effective for establishing field margins with a dense heterogenous plant cover and
434 a flowering continuum spanning the whole crop. As recently reviewed by Gontijo (2019),
435 structurally complex habitats can improve predator survival by providing shelter with a suitable
436 microclimate during summer (cooler temperature and higher humidity) and reduced intraguild
437 predation (either by reducing the chances of direct encounters or by sheltering multiple
438 alternative prey). In addition, the flowering continuum is likely to provide a food resource
439 continuum (nectar, pollen) for natural enemies although the quality and quantity of each
440 constitutive component might be different. In our case, cornflower appeared particularly
441 interesting. First, its abundant flowering spanned the entire melon cropping period. Second, in
442 addition to floral nectar and pollen, it has extrafloral nectaries (Weber et al., 2015) accessible
443 to insects such as syrphids and lacewings (Gilbert, 1981; Limburg and Rosenheim, 2001).
444 Moreover, it hosts a specific aphid *Uroleucon jaceae*, which is harmless to melon crops. Large
445 colonies of this aphid developed during the vegetative growth of the cornflower and attracted
446 numerous specialist predators such as coccinellids (Figure 1b). Such alternative preys and/or
447 their honeydew allow predator populations to build up prior to melon planting thus enhancing
448 the chances of success for biological regulation (Gontijo, 2019).
449 Three complementary trapping methods were used to monitor generalist and specialist
450 predators: pitfall traps and vacuum sampling to assess the attractiveness of the different field

451 margins and the melon crop itself, and interception traps to assess the movement of predators
452 from the margins to the crop. The abundance of spiders assessed by pitfall trapping was highly
453 dependent on the vegetation type. There were respectively 1.6 and 3.2 times more spiders
454 caught in grass and flower margins than in bare soil. Other authors previously observed that
455 bare soil hosted fewer spiders than grassy zones in olive groves (Paredes et al., 2013) and
456 vineyards (Costello and Daane, 1998) and that flower strip implementation resulted in even
457 higher abundances than grass margins (Fernandez et al., 2008; Marko and Keresztes, 2014;
458 Samu, 2003). Within the melon crop, on the contrary, the effect of field margin management
459 was not significant. Spiders concentrated within the flower and grass strips and did not seem to
460 spread much within the melon crop. These results are in agreement with those of Ditner et al.
461 (2013) who observed that, despite high spider abundance in flower margins, their abundance
462 was low in cabbage fields at two distances from the flower margins. This low dispersion of
463 spiders from the field margins towards the crop may be due to both a low attractiveness of the
464 crop or a high attractiveness of the field margins. The vegetative strata within the flower strips
465 may be more suitable (shelter and microclimatic effects) and/or contain more potential prey
466 (Gontijo, 2019). This high attractiveness of the field margins was also obvious for other
467 generalist predators and specialist predators caught by vacuuming. Generalist predators
468 (including spiders) and aphid specialist predators were respectively 5.5 and 9.1 times more
469 abundant in flower margins than in bare soil. Canopy structure and composition, especially the
470 presence of flowers, appear to be the main drivers of generalist and specialist predator
471 populations as observed in other agro-ecological contexts (Gontijo et al., 2013; Mansion-
472 Vaquié et al., 2017). For syrphids, flowers are complementary food resources which, in addition
473 to aphid prey, are necessary to complete their life-cycle: larvae are aphidophagous but adults
474 feed on pollen and nectar (Gilbert, 1981). Nectar sugars are fuel for flight and pollen proteins
475 are necessary for egg maturation. For omnivorous coccinellids and lacewings, flowers are

476 substitutable resources since larvae and adults consume both plant material and prey. For
477 instance, Robinson et al. (2008) observed that the presence of buckwheat flowers enhanced the
478 longevity and fecundity of the lacewing *Micromus tasmaniae* when aphid prey was low. In our
479 study, specialist predators collected by vacuum sampling were mostly coccinellids (91%),
480 rarely lacewings (8%) and syrphids (1%). Lacewings are mainly nocturnal (Vas et al., 1999)
481 whereas samplings were made in the morning explaining their low representativeness. Syrphids
482 larvae also show nocturnal behavior and adults may have easily escaped vaccum sampling due
483 to their fast flight. The effect of flower strips on lacewing and syrphid abundance is therefore
484 better assessed through interception trapping. This third trapping method allowed us to assess
485 arthropods moving from the margins to the crop. Fluxes of generalist and specialist predators
486 moving from flower strips to the melon crop were significantly increased compared to bare soil.
487 Manipulating floral resources was particularly beneficial for coccinellids and syrphids. Other
488 studies reported enhanced abundances of these natural enemies in vegetables cultivated with
489 flower strips. Ribeiro and Gontijo (2017) showed that intercropping alyssum (*Lobularia*
490 *maritima* L.) with collards (*Brassica oleracea* L.) contributed to increase the abundance of
491 generalist predators, including coccinellids and syrphids, which translated into a significant
492 reduction in collard pests, especially aphids. Similarly, White et al. (1995) showed that sowing
493 phacelia around the borders of cabbage crops significantly increased syrphid populations and
494 decreased aphid populations. Using field cages, Hogg et al. (2011) showed that the presence of
495 alyssum enhanced aphid suppression in lettuce through increased syrphid fecundity.
496 Investigating the optimal spatial distribution of floral resources, Gillespie et al. (2011)
497 confirmed that adult syrphids were active dispersers and that aphids were suppressed up to 50
498 m away from the nearest alyssum strip.
499 To conclude, our results confirmed that flower strips specifically designed for melon crops
500 attracted significantly more aphid predators than grass margins and bare soil, in agreement with

numerous previous studies. Their potential value to promote biocontrol was therefore established. However, the difficult question is whether these natural enemies did migrate in the adjacent crop so that pest regulation can occur. Pitfall traps and vacuum samplings performed within the melon crop both suggested that migration was limited. Results from interception traps, however, indicated enhanced coccinellid and syrphid migration fluxes near flower strips compared to bare soil, suggesting a possible dispersion further in the crop. Whether this potential for regulation resulted in an effective impact on aphid infestation and virus epidemics in melon crops remains unanswered. The fact remains that, like any other pest management strategy, habitat management is not a silver bullet and should be combined with other methods in an integrated pest management program. For melon crops, combining sown flower strips with *Vat* gene resistance to *A. gossypii* and the viruses they carry (Boissot et al., 2016; Schoeny et al., 2017) could be an innovative option worth investigating.

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Declarations of interest: none

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## Figure captions

**Figure 1** Flower strip composed of five plant species (cornflower, grass pea, sainfoin, salad burnet and sweet marjoram) selected to boost biological control in melon crop (a) Interception trap used to assess the fluxes of aphid predators from the field margin to the crop (b) Adult coccinellid feeding in a colony of *Uroleucon jaceae* on cornflower. Photo credit: Alexandra Schoeny, INRA

**Figure 2** Abundance of spiders assessed by pitfall trapping, within field margins and melon crop, for three types of field margin management, in a field experiment conducted in Avignon. Pitfalls were placed for one week of monitoring at two dates in 2013 and 2014. Data are means across replicates and dates. Bars show standard errors of the mean. Modalities with same letters are not significantly different according to the post hoc multiple comparison test performed (Tukey HSD, alpha = 0.05).

**Figure 3** Abundance of generalist predators (a) and aphid specialist predators (b) assessed by vacuum sampling, within field margins and melon crop, for three types of field margin management, in a field experiment conducted in Avignon. Samples were collected at two dates in 2012, 2013 and 2014. Data are means across replicates and dates. Bars show standard errors of the mean. Modalities with same letters are not significantly different according to the post hoc multiple comparison test performed (Tukey HSD, alpha = 0.05).

688 **Figure 4** Fluxes of aphid predators migrating from the field margins to the melon crop assessed
689 by interception trapping, for two types of field margin management, in a field experiment
690 conducted in Avignon. Monitoring was carried out for five weeks in 2014 and six weeks in
691 2015. Data are means across replicates and weeks. Bars show standard errors of the mean.
692 Modalities with same letters are not significantly different according to the Wilcoxon signed-
693 rank test performed (alpha = 0.05).

## Table captions

**Table 1** Evaluation of the capacity of 20 plant species to host *Aphis gossypii* and *Myzus persicae*. The acceptance rate (percentage of aphids recovered after 24h) and the reproductive rate (number of offspring per recovered aphid) were assessed in no-choice settling tests conducted under controlled conditions.

**Table 2** Evaluation of the capacity of 19 plant species to host four viruses frequently observed on melon crops in France. The transmission rate (percentage of infected plants) was assessed through aphid transmission (CABYV) or mechanical inoculation (CMV, WMV, ZYMV) under controlled conditions.

**Table 3** Kinetics of flowering for five plant species composing flower strips evaluated in a field experiment conducted in Avignon between 2011 and 2015. For each plant species, the percentage of plants at flowering stage was monitored at different dates after melon planting. Plants were considered at flowering stage when displaying at least one open flower. Data are means ± SEM.

## Highlights

- We selected five plant species minimizing aphid and virus risks in melon.
- We assessed the seed mix in a five-year field experiment.
- Sown flower strips displayed a flowering continuum conducive to natural enemies.
- Generalist and specialist predators were significantly enhanced in flower strips.

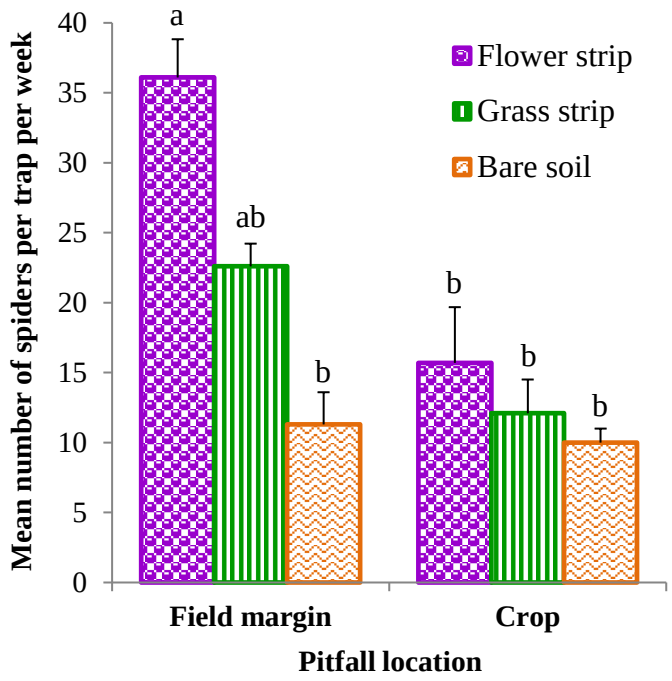
717 **Figure 1**

718 

719 **Figure 2**

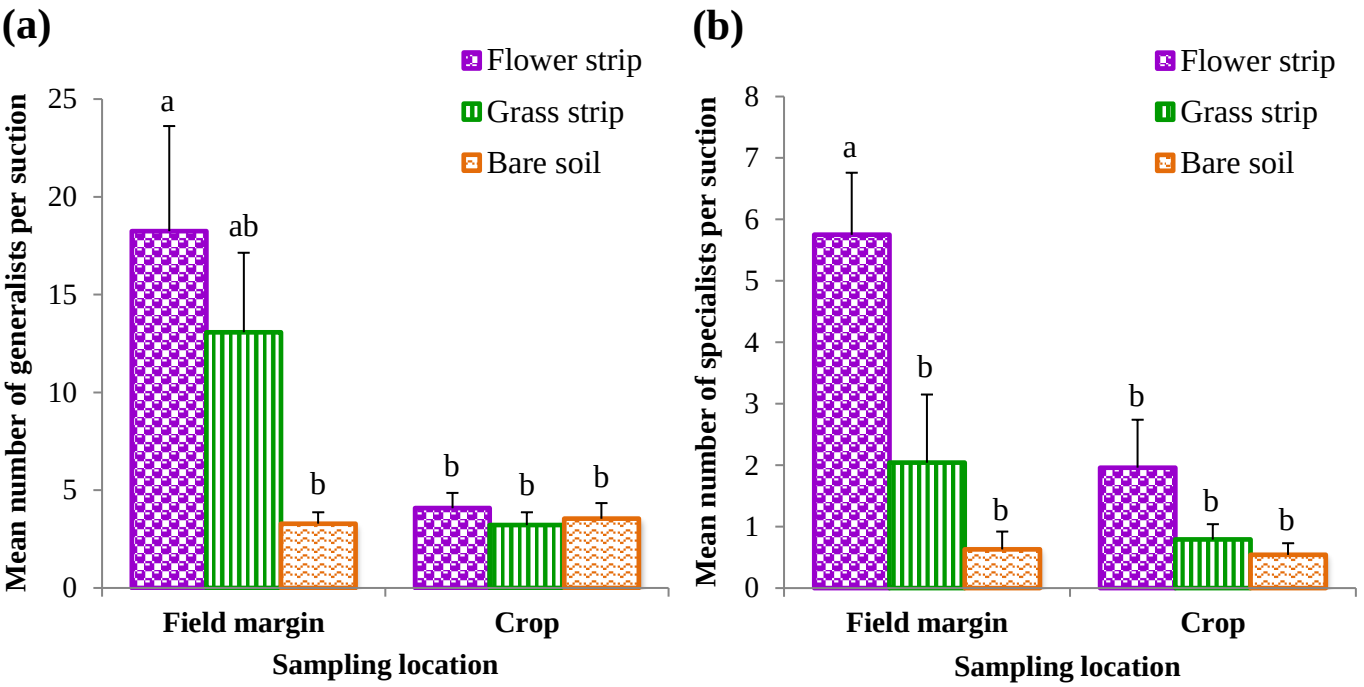
720

721



722 **Figure 3**

723



724 **Figure 4**

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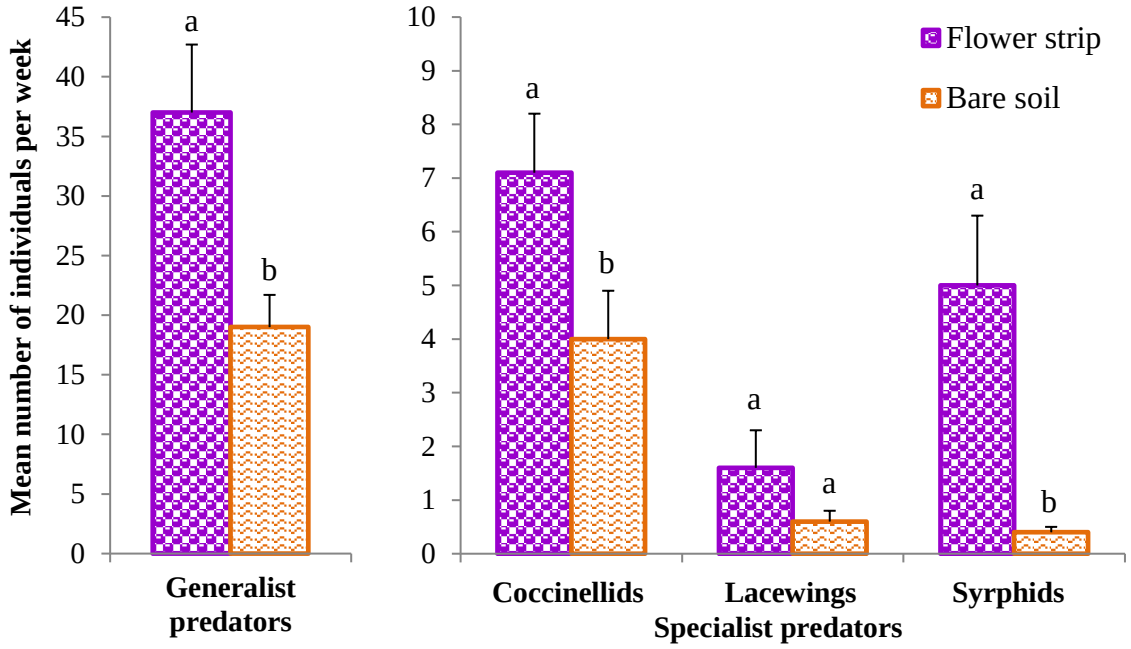


729 **Table 1**

| | *Aphis gossypii* | | | | | | | *Myzus persicae* | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | % aphids recovered after 24h | | | Nb offspring per recovered aphid | | | | % aphids recovered after 24h | | | Nb offspring per recovered aphid | | |
| **Common name** [a] | **Nb tested plants** | **Mean** | **SEM** | **Group** [b] | **Mean** | **SEM** | **Group** [b] | **Nb tested plants** | **Mean** | **SEM** | **Group** [b] | **Mean** | **SEM** | **Group** [b] |
| Basil | 10 | 83 | 6 | ab | 0.0 | 0.0 | c | 10 | 19 | 4 | def | 0.0 | 0.0 | d |
| Borage | 10 | 24 | 6 | defgh | 7.2 | 1.8 | ab | 10 | 95 | 3 | a | 4.1 | 0.3 | ab |
| Buckwheat | 10 | 3 | 2 | h | 2.4 | 2.3 | bc | 10 | 0 | 0 | f | 0.0 | 0.0 | d |
| Bullwort | 10 | 74 | 6 | abc | 1.5 | 0.3 | abc | 10 | 90 | 4 | ab | 1.1 | 0.4 | bcd |
| Corn marigold | 10 | 4 | 2 | h | 2.6 | 1.3 | bc | 10 | 74 | 7 | abc | 2.6 | 0.3 | abcd |
| Cornflower * | 30 | 67 | 7 | bc | 1.4 | 0.2 | bc | 30 | 71 | 6 | bc | 1.6 | 0.4 | bcd |
| Dill | 10 | 12 | 2 | fgh | 0.1 | 0.1 | c | 10 | 77 | 8 | abc | 1.5 | 0.4 | abcd |
| Field marigold | 10 | 57 | 7 | bcd | 0.8 | 0.2 | bc | 10 | 97 | 3 | a | 1.1 | 0.1 | abcd |
| French marigold | 10 | 71 | 4 | abc | 0.0 | 0.0 | c | 10 | 31 | 3 | de | 0.2 | 0.1 | bcd |
| Grass pea * | 20 | 10 | 3 | gh | 0.8 | 0.3 | bc | 20 | 28 | 5 | de | 1.0 | 0.3 | bcd |
| Lacy phacelia | 10 | 57 | 7 | bcd | 18.4 | 1.3 | a | 10 | 48 | 11 | cd | 4.0 | 0.9 | abc |
| Marigold | 10 | 21 | 5 | efgh | 0.4 | 0.3 | bc | 10 | 44 | 6 | cd | 0.1 | 0.1 | cd |
| Melon ** | 103 | 95 | 1 | a | 12.0 | 0.6 | a | 10 | 2 | 2 | f | 0.1 | 0.1 | d |
| Nigella | 10 | 1 | 1 | h | 0.0 | 0.0 | c | 10 | 40 | 10 | d | 0.8 | 0.2 | bcd |
| Pepper ** | nt | nt | nt | nt | nt | nt | nt | 94 | 94 | 1 | a | 6.0 | 0.2 | a |
| Ryegrass | 20 | 32 | 8 | def | 0.0 | 0.0 | c | 20 | 9 | 3 | ef | 0.0 | 0.0 | d |
| Sainfoin * | 20 | 29 | 8 | defg | 0.0 | 0.0 | c | 20 | 7 | 4 | f | 0.0 | 0.0 | d |
| Salad burnet * | 20 | 9 | 2 | gh | 0.2 | 0.1 | c | 20 | 5 | 2 | f | 0.1 | 0.1 | d |
| Sweet marjoram * | 20 | 49 | 11 | cde | 0.0 | 0.0 | c | 20 | 32 | 7 | d | 0.1 | 0.1 | d |
| White campion | 10 | 10 | 4 | fgh | 0.0 | 0.0 | c | 10 | 30 | 6 | de | 0.0 | 0.0 | d |
| **Statistical test** | | | | Chi2 | | | KW | | | | Chi2 | | | KW |
| **P-value** | | | | < 0,0001 | | | < 0,0001 | | | | < 0,0001 | | | < 0,0001 |

730 nt= not tested

731 [a] Plant species with an asterisk are included in the flower mix

732 Plant species with two asterisks are reference species:

733 Melon is reference species for *Aphis gossypii* risk assessment

734 Pepper is reference species for *Myzus persicae* risk assessment

735 [b] Plants with the same letters are not significantly different

736 Grey zones highlight acceptance and reproductive rates not significantly different to the reference rates obtained on plant species used for aphid rearing

737 **(**melon for *A. gossypii* and pepper for *M. persicae*).

738 **Table 2**

739

| | Virus (isolate) | | | | | |
|---|---|---|---|---|---|---|
| | **CABYV (FIP)** | **CMV (14)** | **WMV (LL1A)** | **WMV (LL2B3)** | **ZYMV (E9)** | **ZYMV (124L11)** |
| **Common name [a]** | **Nb infected plants / nb tested plants [b]** | | | | | |
| Basil | 0/8 b | 6/8 ab | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Borage | 0/8 b | 7/8 a | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Buckwheat | 0/8 b | 14/16 a | 10/16 ab | 11/16 a | 0/8 b | 0/8 b |
| Bullwort | 0/8 b | 0/8 b | 8/8 a | 8/8 a | 8/8 a | 8/8 a |
| Corn marigold | 0/8 b | 6/6 a | 0/6 b | 0/6 b | 0/6 b | 0/6 b |
| Cornflower * | 0/8 b | 13/16 a | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Dill | 3/8 a | 0/8 b | 8/8 a | 8/8 a | 8/8 a | 8/8 a |
| Field marigold | 0/8 b | 8/8 a | 8/8 a | 8/8 a | 0/8 b | 0/8 b |
| French marigold | 0/8 b | 8/8 a | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Grass pea * | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Lacy phacelia | 0/8 b | 8/8 a | 6/6 a | 6/6 a | 0/6 b | 0/6 b |
| Marigold | 0/8 b | 8/8 a | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Melon | 36/100 a | 27/28 a | 27/28 a | 27/28 a | 20/20 a | 20/20 a |
| Nigella | 0/8 b | 8/8 a | 8/8 a | 8/8 a | 8/8 a | 8/8 a |
| Ryegrass | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Sainfoin * | 0/8 b | 0/6 b | 0/6 b | 0/6 b | 0/6 b | 0/6 b |
| Salad burnet * | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b | 0/8 b |
| Sweet marjoram * | 0/8 b | 7/8 a | 0/7 b | 0/7 b | 0/7 b | 0/7 b |
| White campion | 0/8 b | 5/6 a | 0/6 b | 0/6 b | 0/6 b | 0/6 b |
| **Statistical test** | Chi2 | Chi2 | Chi2 | Chi2 | Chi2 | Chi2 |
| ***P*-value** | < 0,0001 | < 0,0001 | < 0,0001 | < 0,0001 | < 0,0001 | < 0,0001 |

740

741 [a] Plant species with an asterisk are included in the flower mix

742 [b] Plants with the same letters are not significantly different

743 Grey zones highlight transmission rates not significantly different to the reference rates obtained on

744 melon.

745 **Table 3**

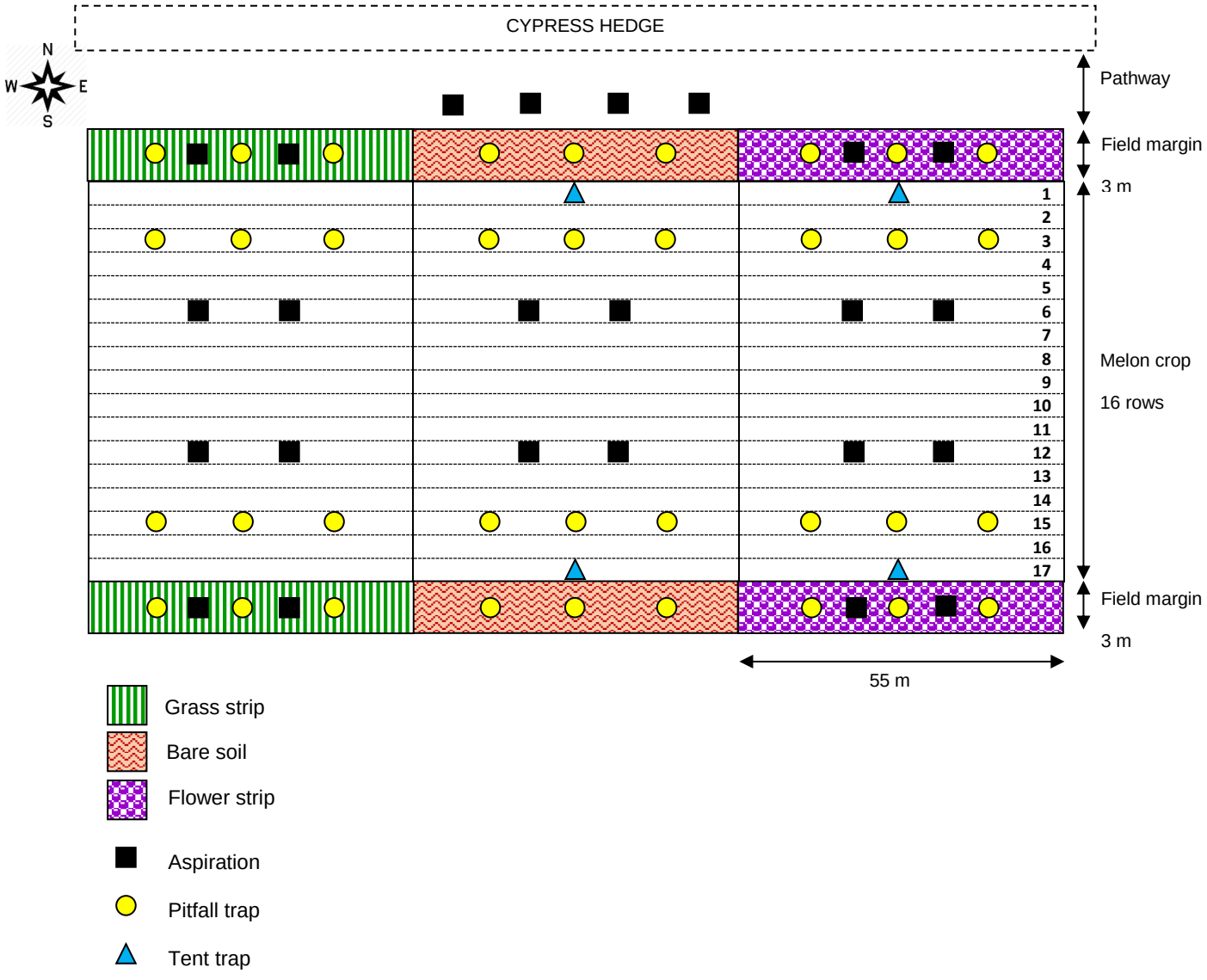
| | | Days after melon planting | | | | | |
|---|---|---|---|---|---|---|---|
| **Common name** | **Scientific name** | **[1-10]** | **[11-20]** | **[21-30]** | **[31-40]** | **[41-50]** | **[51-60]** |
| Cornflower | *Centaurea cyanus* | 12±12 | 56±14 | 71±8 | 87±6 | 73±7 | 34±6 |
| Grass pea | *Lathyrus sativus* | 82±10 | 97±3 | 83±11 | 19±9 | 2±1 | 0±0 |
| Sainfoin | *Onobrychis viciifolia* | 1±1 | 21±7 | 43±6 | 45±9 | 50±14 | 42±3 |
| Salad burnet | *Sanguisorba minor* | 0±0 | 8±3 | 31±8 | 20±7 | 21±10 | 39±7 |
| Sweet marjoram | *Origanum majorana* | 0±0 | 0±0 | 0±0 | 2±2 | 28±19 | 2±2 |

746

## Graphical abstract

Supplementary material

**Figure S1** Schematic representation of the experimental design with spatial pattern of arthropod samplings in field margins and melon crop in 2014



Supplementary material

**Table S1** Biotests conducted in controlled conditions to evaluate the capacity of 20 plant species to host two aphid species and and four viruses frequently observed on melon crops in France

| Common name [a] | Scientific name | Botanical family | Nb of weeks after sowing | Aphid risks [c] | | Virus risks [d] | | Global risks [e] |
|---|---|---|---|---|---|---|---|---|
| | | | | Biotests | Literature | Biotests | Literature | |
| Basil | *Ocimum basilicum* L. | Lamiaceae | 6-7 | Ag | Ag, Mp | C | C, W | Ag, Mp, C, W |
| Borage | *Borago officinalis* L. | Boraginaceae | 4 | Ag, Mp | Ag | C | C | Ag, Mp, C |
| Buckwheat | *Fagopyrum esculentum* Moench | Polygonaceae | 4 | ∅ | Ag, Mp | C, W | C | Ag, Mp, C, W |
| Bullwort | *Ammi majus* L. | Apiaceae | 5 | Ag, Mp | Mp | W, Z | C, W, Z | Ag, Mp, C, W, Z |
| Corn marigold | *Chrysanthemum segetum* L. | Asteraceae | 5 | Mp | Mp | C | C | Mp, C |
| Cornflower * | *Centaurea cyanus* L. | Asteraceae | 4-5 | ∅ | ∅ | C | C | C |
| Dill | *Anethum graveolens* L. | Apiaceae | 5-6 | Mp | Ag, Mp | CA, W, Z | C | Ag, Mp, CA, C, W, Z |
| Field marigold | *Calendula arvensis* L. | Asteraceae | 3-5 | Mp | Ag, Mp | C, W | C | Ag, Mp, C, W |
| French marigold | *Tagetes patula* L. | Asteraceae | 6 | Ag | Ag, Mp | C | C | Ag, Mp, C |
| Grass pea * | *Lathyrus sativus* L. | Leguminosae | 4 | ∅ | ∅ | ∅ | C | C |
| Lacy phacelia | *Phacelia tanacetifolia* Benth. | Hydrophyllaceae | 4 | Ag, Mp | Mp | C, W | C, W | Ag, Mp, C, W |
| Marigold | *Calendula officinalis* L. | Asteraceae | 5 | ∅ | Ag, Mp | C | C | Ag, Mp, C |
| Melon ** | *Cucumis melo* L. | Cucurbitaceae | 3-5 | Ag | Ag, Mp | CA, C, W, Z | CA, C, W, Z | Ag, Mp, CA, C, W, Z |
| Nigella | *Nigella damascena* L. | Ranunculaceae | 5 | ∅ | ni | C, W, Z | ni | C, W, Z |
| Pepper ** | *Capsicum annuum* L. | Solanaceae | 5-8 | na | na | na | na | na |
| Ryegrass | *Lolium perenne* L. | Poaceae | 7-8 | ∅ | Mp | ∅ | ni | Mp |
| Sainfoin * | *Onobrychis viciifolia* Scop. | Leguminosae | 5-7 | ∅ | ∅ | ∅ | ni | ∅ |
| Salad burnet * | *Sanguisorba minor* Scop. | Rosaceae | 5-6 | ∅ | ∅ | ∅ | ni | ∅ |
| Sweet marjoram * | *Origanum majorana* L. | Lamiaceae | 6-8 | ∅ | ∅ | C | ni | C |
| White campion | *Silene latifolia* Poir. | Caryophyllaceae | 6 | ∅ | Mp | C | C | Mp, C |

na= not applicable
ni= no information available

[a] Plant species with an asterisk are included in the flower mix
Plant species with two asterisks are reference species:
Melon is reference species for *Aphis gossypii* and virus risk assessment
Pepper is reference species for *Myzus persicae* risk assessment

[b] Plants with the same letters are not significantly different

[c] Aphid risks according to Blackman and Eastop 2006, Kavallieratos et al., 2007
Ag (Aphis gossypii)
Mp (Myzus persicae)

[d] Virus risks according to:
CA (CABYV): Lecoq et al., 1992
C (CMV): Edwardson and Christie, 1997
W (WMV): Edwardson and Christie, 1991
Z (ZYMV): Desbiez and Lecoq, 1997

[e] Underlined risks correspond to new descriptions compared to literature
∅= risk considered as negligible according biotests and/or literature