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**Can insectary plants enhance the presence of natural enemies of the green peach  
aphid (Hemiptera: Aphididae) in Mediterranean peach orchards?**

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21 **Abstract**

22 Conservation biological control could be an alternative to insecticides for the manage-  
23 ment of the aphid *Myzus persicae* (Sulzer). To develop sustainable strategies for *M. per-*  
24 *sicae* control in peach orchards in the Mediterranean, a two-year field experiment was  
25 conducted to identify the key predators of the aphid; to determine whether the prox-  
26 imity of insectary plants boost natural enemies of *M. persicae* in comparison to the res-  
27 ident vegetation; and whether selected insectary plants enhance natural enemy popu-  
28 lations in the margins of peach orchards. *Aphidoletes aphidimyza* Rondani and *Episyr-*  
29 *phus balteatus* De Geer were the most abundant predators found among sentinel aphid  
30 colonies, accounting for 57% and 26%, respectively. Samplings during 2015 yielded twice  
31 as many hoverflies in *M. persicae* sentinel plants close to the insectary plants as those  
32 close to the resident vegetation. The abundance of other natural enemies in sentinel  
33 plants, depending on their proximity to the insectary plants, was not significantly differ-  
34 ent in either of the two years. Hoverflies hovered more often over the insectary plants  
35 than over the resident vegetation and landed significantly more often on *Lobularia mar-*  
36 *itima*, *Moricandia arvensis*, and *Sinapis alba* than on *Achillea millefolium*. Parasitoids  
37 were significantly more abundant in *L. maritima* and *A. millefolium*. The vicinity of se-  
38 lected insectary plants to peach orchards could improve the presence of hoverflies,  
39 which might benefit the biological control of *M. persicae*.

40

41 **Keywords**

42 Conservation biological control, agroecological infrastructures, insectary plants, parasi-  
43 toids, predators.

44

## 45 INTRODUCTION

46 Peach (*Prunus persica* [L.] Batsch) is an important crop in Europe, the second worldwide  
47 producer after China. Within Europe, Spain is a lead producer, with 30% of total Euro-  
48 pean production (FAOSTAT 2017). Catalonia comprises 24% of the total Spanish produc-  
49 tion, mostly in the province of Lleida, where 20,000 ha are dedicated to this crop (MAPA  
50 2017).

51 The green peach aphid, *Myzus persicae* (Sulzer) (Hemiptera: Aphididae), is one of the  
52 most important pests of peach, one of its primary hosts. Although to our knowledge  
53 there has been no formal evaluation of yield loss in peach production due to this aphid  
54 species, it has been acknowledged as a very injurious pest (Dedryver et al. 2010). Dam-  
55 ages to peach include leaf twisting, pitting and discolored fruits, and the vectoring of  
56 important viruses, such as plum pox, also known as sharka (Penvern et al. 2010, Bar-  
57 bagallo et al. 2017).

58 Aphids in peach crops have usually been managed with insecticide sprays (Barbagallo et  
59 al. 2017). However, the use of pesticides is a growing social concern due to the risks that  
60 these products pose to human health and to the environment. Additionally, *M. persicae*  
61 resistance to numerous active substances, such as pyrethroids, neonicotinoids, organo-  
62 phosphates, and carbamates, often renders many insecticide treatments ineffective  
63 (Foster et al. 2011). Therefore, it is urgent to develop alternative aphid management  
64 strategies that are more sustainable and socially acceptable. In this scenario, biological  
65 control, more specifically conservation biological control, could be a good tool to reduce

66 the use of insecticides in peach crops (Dedryver et al. 2010, Penvern et al. 2010). Con-  
67 servation biological control relies on modifying the environment or existing practices to  
68 protect and enhance specific natural enemies or other organisms to reduce the effect  
69 of pests on crops (Eilenberg et al. 2001).

70 Aphid natural enemies belong to different taxonomic groups from entomopathogenic  
71 organisms to parasitoids and also include several specialist and generalist predators.  
72 Among these predators, those belonging to the Cecidomyiidae, Coccinelliidae, Chrysopi-  
73 dae, and Syrphidae families are the most commonly mentioned in the literature  
74 (Brodeur et al. 2017). There are few studies about the aphid natural enemies present in  
75 peach orchards in the Mediterranean (Remaudière and Leclant 1971, Penvern et al.  
76 2010, Aparicio et al. 2019). In Spain, Avilla et al (2008) stated that the rich complex of  
77 aphid natural enemies present in peach orchards, including parasitoids and generalist  
78 predators, are not enough to provide satisfactory pest control. However, these authors  
79 did not explicitly state which natural enemies were present or their relative abundance.

80 The intensification of agriculture, the loss of habitat, and the use of insecticides have  
81 caused a lack of adequate resources for beneficial insects, including the natural enemies  
82 of aphids, in agricultural ecosystems (Landis et al. 2000, Biesmeijer et al. 2006, Haaland  
83 et al. 2011). Some strategies to overcome this scenario and to enhance the presence of  
84 beneficial insects in crops often involve the addition of floral resources at the farm level  
85 (Kremen and Chaplin-Kramer 2007). These floral resources provide nectar, pollen, and  
86 shelter to natural enemies when the crop is not in bloom (Lavandero et al. 2005, Hogg  
87 et al. 2011) and contribute to enabling that the complex of predators and parasitoids  
88 remains in the agroecosystem (Wyss 1996, Brown and Schmitt 2001). The presence of

89 ecological infrastructures with flowering plants in orchards has already been shown to  
90 enhance the populations of some aphid species' natural enemies (Tylianakis et al. 2004,  
91 Miñarro et al. 2005, Gontijo et al. 2013, Rodríguez-Gasol et al. 2019). Nonetheless,  
92 flower species must be carefully chosen, considering that they must be accessible to  
93 natural enemies, they must be adapted to the environment, and they will not be a res-  
94 ervoir of pests or diseases harmful to the crop (Jervis et al. 1993, Baggen and Gurr 1998,  
95 Hogg et al. 2011).

96 With the aim of developing sustainable aphid management strategies based on conser-  
97 vation biological control, the objectives of this study were: 1) to identify the key preda-  
98 tors of *M. persicae* in peach orchards in Spain's Mediterranean fruit production area; 2)  
99 to determine whether the proximity of the selected insectary plants to the crop may  
100 boost the abundance of natural enemies of *M. persicae*, and 3) to determine whether  
101 insectary plants, previously identified as promising candidates, enhance natural enemy  
102 populations in the margins of peach orchards and to assess whether they harbor pests  
103 harmful to the crop.

104

## 105 **MATERIALS AND METHODS**

### 106 Study area and experimental setup

107 The study was conducted in 2015 and 2016 in four organic peach orchards located in the  
108 area of Segrià (Lleida, Catalonia) in the northeast of Spain. All sampled fields were under  
109 supervision of the same pest management advisor, with minimal use of pesticides. In  
110 each orchard, four patches of 1 m<sup>2</sup> of sown insectary plants (hereafter "sown flower

111 patches”) were planted in a row in one of the field margins about 5 m from the first row  
112 of trees (Fig. 1). Another four patches of 1m<sup>2</sup> of weedy resident vegetation were selected  
113 in another unmowed field margin, also about 5 m from the first row of trees, and were  
114 used as controls. The sown flower patches were 5 m apart, same as the resident vege-  
115 tation patches, and the distances between the sown flower and the resident vegetation  
116 patches varied according to the field shape and size and ranged from 25 to 130 m.

117 Each sown flower patch consisted of four boxes (50 cm length, 35.5 cm width, and 31  
118 cm height), each planted with 10 plants of one of the following insectary plant species:  
119 *Achillea millefolium* L. (Compositae), *Lobularia maritima* L. (Brassicaceae), *Moricandia*  
120 *arvensis* L. (Brassicaceae), or *Sinapis alba* L. (Brassicaceae). These plants were selected  
121 according to our group’s previous results (Arnó et al. 2012, 2018). The plants were grown  
122 from seeds in pots with potting soil in a greenhouse, transplanted to the boxes, and  
123 taken to the field by mid-April in 2015 and by mid-March in 2016. Within each sown  
124 flower patch, position of the different plant species was randomized. They were period-  
125 ically drip-irrigated similarly to the trees, and in both years, the plants were kept in the  
126 sites until mid-September. Most of the plants were taken to the field in bloom, and their  
127 phenological stage during the sampling is compiled in Table 1. The resident vegetation’s  
128 plant composition was highly variable and included several species commonly found in  
129 fruit orchard margins. Table 2 summarizes the species that, while flowering, were pre-  
130 sent in more than 25% of the patches’ surfaces during the samplings.

131

132 Natural enemies associated with *M. persicae*

133 Sentinel plants were used to identify the natural enemies associated with *M. persicae*.  
134 For this, small potted peach plants (two to three years old and approximately 50 cm high  
135 in 2-liter pots) were infested *ad-hoc* with approximately 100 *M. persicae* (adults and  
136 mixed instars) obtained from infested peach shoots collected from the same orchards  
137 in Lleida. The infested sentinel plants were kept for a week in a closed screened green-  
138 house to prevent contamination by other aphid species or colonization by natural ene-  
139 mies and to allow aphid population increase. Afterwards, 16 sentinel plants were taken  
140 to each orchard and placed under the peach canopy. To avoid plant desiccation, they  
141 were placed into bigger pots filled with water. These pots' outsides were sprayed with  
142 insect-trapping glue (Soveurode® aérosol, Plantin SARL, Courthézon, France) to prevent  
143 ants and other soil predators from climbing the plant. The sentinel plants were placed  
144 at two distances from the field edge: four plants were placed next to the first rows of  
145 peaches, just in front of the sown flower patches, and four plants were placed at a dis-  
146 tance of ca. 5m, between the second and the third rows of peaches. Following the same  
147 pattern, another group of eight sentinel plants was placed in front of the resident vege-  
148 tation. The sentinel plants were taken to the orchards four times at 15-day intervals  
149 starting in week 18 (beginning of May) to week 24 (mid-June). They were left there for  
150 one week and then taken to the laboratory, where all the predators were carefully re-  
151 covered regularly during the following four weeks.

152 All Cecidomyiidae larvae present in those shoots were placed on microscope slides and  
153 morphologically classified using Harris' key (Harris 1973) Hoverfly larvae (Diptera: Syr-  
154 phidae) and *Orius* spp. (Hemiptera: Anthocoridae) nymphs and adults were individually  
155 stored at -20°C for subsequent molecular identification by conventional PCR following



156 the methods developed by Gomez-Polo et al. (2014) and Gomez-Polo et al. (2013), re-  
157 spectively. Nonamplified specimens were tested using the universal primers ZBJ-ArtF1c  
158 and ZBJ-ArtR2c (Zeale et al. 2011) to confirm a DNA presence in those extractions, using  
159 the following cycling condition: initial denaturation at 94° C for 5', followed by 40 cycles  
160 of 94° C for 30'', 46° C for 45'', 68° C for 45'', and a final extension of 68° C for 10'. All  
161 PCR products were analyzed by gel electrophoresis (2.4% agarose gels) and visualized  
162 with GelRed® (Biotium, Hayward, CA).

163

#### 164 Natural enemies and phytophagous insects on insectary plants

165 To evaluate the attractiveness of insectary plants to natural enemies and phytophagous  
166 insects, the sown flower and resident vegetation patches were sampled using visual ob-  
167 servations and the beating tray method (hereafter "beating"). For the visual observa-  
168 tions, the number of adult hoverflies hovering above each patch for three minutes was  
169 recorded. In the sown flower patches, the number of hoverflies that landed on each  
170 insectary plant species during the same period was also recorded. The beating targeted  
171 the insect community that cannot be seen during their flight and are usually within the  
172 plant foliage or in the flowers. In the sown flower patches, a sample was obtained by  
173 hand-beating three consecutive times a handful of each plant species separately on a  
174 plastic white tray (24 x 35 cm). Insects that fell onto the tray were visually classified *in*  
175 *situ* as hymenopteran parasitoids, *Orius* spp., ladybeetles, lacewings, aphids, or thrips  
176 other than *Aeolothrips* spp. (hereafter "thrips"). *Aeolothrips* spp. were excluded from  
177 the records because they are not known to be aphid predators but thrips predators (Riu-

178 davets 1995). The same methods and records were conducted on the resident vegeta-  
179 tion, but instead of individual plant species, five and four randomly selected samples  
180 were obtained by beating a handful of vegetation per patch in 2015 and in 2016, respec-  
181 tively. All individuals were returned to the patches after the identification, and all beat-  
182 ings were always done after the visual observations. Both methods were conducted fort-  
183 nightly from week 18 to week 24 in 2015 (end of April to mid-June). Samplings were  
184 done on sunny days (always above 15° C) from 9.00–13.00. Sixteen samples for field and  
185 plant species were obtained from the sown flower patches, and 20 (2015) and 16 (2016)  
186 samples from the resident vegetation.

187

#### 188 Data analysis

189 The data for each year was analyzed separately using a generalized linear mixed-effects  
190 model (GLMM) with a negative binomial (NB) response distribution. The number of in-  
191 sects collected by beating and the number of landings by hoverflies per insectary plant  
192 species in the visual observations were the response variables. The sown flower and  
193 resident vegetation patches (treatment factor) were the main fixed effect. The orchard,  
194 patch, week and the interaction treatment\*week were the random factors. Pairwise  
195 comparisons (post-hoc tests) were carried out using Tukey's method for multiple com-  
196 parisons. The number of arthropods in the beating sampling was compared among five  
197 treatments: the four insectary plants and the resident vegetation. The number of syr-  
198 phid adults' landings was compared among the four insectary plants. Data from the vis-  
199 ual observations were referred to as number of hoverflies per patch (1m<sup>2</sup>) and per in-  
200 sectary plant species and time (3 minutes). Arthropods in the beating and in the sentinel

201 plants were referred to as individuals per white tray and per sentinel plant, respectively.  
202 Abundances of natural enemies in the sentinel plants placed at 0 and 5 m within the  
203 orchard (Fig. 1) were jointly analyzed since a preliminary analysis did not show signifi-  
204 cant differences between the two distances. The statistical analysis was performed using  
205 R v3.5.3. A nominal significance level of 5% ( $P < 0.05$ ) was applied in all statistical tests.

206

## 207 **RESULTS**

208 Key predators of *M. persicae* in peach orchards in the Mediterranean area

209 Cecidomyiidae and Syrphidae larvae were the most abundant predators found in the  
210 sentinel plants and represented 57% and 26%, respectively, of the total number of pred-  
211 ators collected during both years (Table 3). The morphological identification of the col-  
212 lected Cecidomyiidae yielded a single species, namely *Aphidoletes aphidimyza* Rondani.  
213 It was consistently present during both years, and its population peaked on week 22  
214 (end of May, Fig. 2a). Regarding hoverflies, 83 larvae were collected in 2015, 60 were  
215 identified by multiplex PCR, 55 were *Episyrphus balteatus* De Geer, and the other five  
216 were *Sphaerophoria* spp. (Le Peletier & Serville). The remaining 23 larvae did not show  
217 amplification with the syrphid-specific primers used. In 2016, 19 hoverfly larvae were  
218 collected, seven larvae identified as *E. balteatus* and three as *Sphaerophoria* spp. The  
219 remaining 9 syrphids did not show amplification. Nevertheless, all non-amplified speci-  
220 mens from both years were amplified using the universal primers as positive controls,  
221 indicating that they might be other syrphid species. The same phenological pattern was  
222 observed in both years, with populations peaking in week 20 (mid-May) (Fig. 2b).

223 Adults and immatures of *Orius* spp., ladybeetles, and lacewings were less common (Ta-  
224 ble 3). In 2015, 32 *Orius* spp., immatures and adults, were collected, but none in 2016.  
225 The conventional PCR allowed the identification of 21 *Orius majusculus* Reuter and three  
226 *Orius niger* (Wolff) (Hemiptera: Anthocoridae). The remaining eight *Orius* individuals did  
227 not show amplification with the primers used; however, amplification with the universal  
228 primers, indicated that they might be other anthocorid species. Most of the *Orius* spp.  
229 were found at the end of April and during the end of May.

230

231 Proximity of sown flower patches on natural enemies of *M. persicae*

232 In regards to the proximity to the sown flower patches or resident vegetation, significant  
233 differences were only found for hoverfly larvae in 2015. In this year, the number in the  
234 sentinel plants near flower patches was twofold higher than in sentinel plants near res-  
235 ident vegetation (Table 4).

236

237 Insectary plants as resources for natural enemies and as reservoirs for peach pests

238 The number of hoverflies hovering over the sown flower patches (Fig. 3) was significantly  
239 higher than that over the resident vegetation (2015:  $Z=6.130$ ,  $P<0.0001$ ; 2016:  $Z=4.111$ ,  
240  $P<0.0001$ ). In 2015 and 2016, hoverflies landed significantly more times on *L. maritima*,  
241 *M. arvensis*, and *S. alba* than on *A. millefolium* (2015:  $\chi^2=24.91$ ,  $P<0.0001$ ; 2016:  
242  $\chi^2=21.14$ ,  $P<0.0001$ ) (Fig. 4).

243 The results obtained via beating indicate that in the sown flower and resident vegetation  
244 patches, the most abundant natural enemies were hymenopteran parasitoids (59% and

245 45% of the total in 2015 and 2016, respectively), followed by polyphagous predators  
246 such as ladybeetles, *Orius* spp., and lacewings (74, 49, and 12 individuals, respectively,  
247 pooling together the data from 2015 and 2016). In addition to potential natural enemies,  
248 a total of 1165 aphids and 2357 thrips were counted during 2015, while 531 aphids and  
249 875 thrips were counted in 2016.

250 In both years, significantly higher numbers of hymenopteran parasitoids were found on  
251 *L. maritima* and *A. millefolium* than in the resident vegetation (2015:  $\chi^2=50.650$ ,  
252  $P<0.0001$ ; 2016:  $\chi^2=22.323$ ,  $P=0.0001$ ) (Fig. 5a). The abundance on *M. arvensis* and on  
253 *S. alba* had values similar or lower to that on the resident vegetation.

254 In 2015, significantly higher numbers of Coccinellidae were recorded in *L. maritima* and  
255 *S. alba* compared to the resident vegetation ( $\chi^2=13.975$ ,  $P=0.0073$ ) (Fig. 5b). In 2016,  
256 ladybeetles were only found on *L. maritima* and on resident vegetation. No statistical  
257 analysis was performed for the 2016 data due to the absence of these predators in most  
258 of the insectary plants. No significantly different *Orius* spp. populations were found in  
259 the insectary plants and the resident vegetation (2015:  $\chi^2=6.052$ ,  $P=0.1952$ ; 2016:  
260  $\chi^2=7.6798$ ,  $P=0.10404$ ) (Fig. 5c). However, in 2015, the highest number of individuals  
261 was recorded in *L. maritima*. Due to the low number of lacewings (2 and 10 in 2015 and  
262 2016, respectively), no statistical analysis was performed.

263 Aphids were recorded in all insectary plants and in the resident vegetation (Fig. 6a).  
264 However, abundances were different among different plant species (2015:  $\chi^2=39.086$ ,  
265  $P<0.0001$ ; 2016:  $\chi^2=19.554$ ,  $P<0.0001$ ). Although these aphids were not identified, our  
266 visual inspections revealed that they were neither *M. persicae* nor *Hyalopterus* spp. In  
267 both years, the number of thrips on *S. alba* was significantly higher than in *L. maritima*

268 and the resident vegetation (2015:  $\chi^2=24.614$ ,  $P<0.0001$ ; 2016:  $\chi^2=16.150$ ,  $P=0.0028$ )  
269 (Fig. 6b).

270

## 271 **DISCUSSION**

272 Our results showed that although five different groups of predators were found on the  
273 sentinel aphid colonies, the gall-midge *A. aphidimyza* was the most abundant followed  
274 by the hoverfly *E. balteatus*. Sporadic observations on *M. persicae* infesting trees were  
275 made on the same fields, and the guild of natural enemies present coincided with those  
276 reported in this study (author's unpublished). These two groups of predators have been  
277 recognized as present in aphid colonies in other Mediterranean orchards, such as in  
278 peach trees in the southeast of France (Remaudière and Leclant 1971) and in apple trees  
279 located in the same area of our study (Rodríguez-Gasol et al. 2019).

280 The abundance of *E. balteatus* above other hoverfly species as aphid predators in fruit  
281 orchards may be related to seasonality, because *M. persicae* attacks peach trees early  
282 in the season when temperatures are still moderate. *Episyrphus balteatus* is not well  
283 adapted to high temperatures (above 25° C), and high rates of mortality occur when this  
284 temperature is exceeded (Hart and bale 1997). Conversely, *Sphaerophoria* spp. is better  
285 adapted to higher temperatures. Since our samplings were carried out during spring, it  
286 may explain the high records of *E. balteatus*.

287 In our samplings, *A. aphidimyza* appeared later in the season. The later presence of *A.*  
288 *aphidimyza* compared to *E. balteatus* has also been reported in apple orchards (Miñarro

289 et al. 2005, Brown and Lightne 1997). These two predators, together with *Aphidius mat-*  
290 *ricariae* Haliday (Hymenoptera: Braconidae), the most abundant parasitoid of *M. persi-*  
291 *cae* in the study area (Aparicio et al. 2019), must be considered the key natural enemies  
292 of *M. persicae* in peach orchards due to their abundance during the period when the  
293 heaviest infestations of *M. persicae* occur.

294 Our surveys also demonstrate that *O. majusculus* and *O. niger* colonized sentinel plants  
295 infested with *M. persicae* and even laid eggs on those plants, since both adults and  
296 nymphs were found on aphid-infested shoots. These two species are different from  
297 *Orius* species recorded by other authors in peach orchards, namely *O. laevigatus* (Fieber)  
298 (Avilla et al. 2008) and *O. minutus* L. (Remaudière and Leclant 1971). These differences  
299 might be influenced by our sampling sites, which were close to other orchards and ara-  
300 ble crops, (e.g., wheat, barley, maize, alfalfa, oats, and rye grass), since *O. majusculus*  
301 has been recorded in these crops in the area, and it has been proven that they migrate  
302 to nearby orchards (Madeira et al. 2014, Ardanuy et al. 2017). The presence of *O. ma-*  
303 *jusculus* in the infested sentinel peach plants might be taken as an indicator of predator  
304 activity in the orchard. This species is a well-known thrips predator (Riudavets and  
305 Castañé 1998), although it has also been described as feeding on aphids (Alvarado et al.  
306 1997, Gómez-Polo et al. 2016). Therefore, their presence can be positive for the biolog-  
307 ical control of both aphids and thrips, which are relevant pests in nectarine cultivars.

308 Surprisingly, other predators such as ladybeetles and lacewings, which are generally rec-  
309 ognized as aphid predators in deciduous fruit trees (Barbagallo et al. 2017), have been  
310 found only in small numbers in the present survey. These findings might be related to  
311 the seasonality of our samplings with the sentinel plants (beginning of May to mid-June),

312 which were conducted mostly during the period when *M. persicae* populations damage  
313 crops in the study area. Miñarro et al. (2005), Dib et al. (2010), and Rodríguez-Gasol et  
314 al. (2019) recorded higher numbers of Coccinellidae and Chrysopidae than our samples  
315 in apple orchards later in the season (beginning of July).

316 The proximity of the sown flower patches to the peach orchards changed the abundance  
317 of hoverflies in 2015, when there were twice as many individuals in the sentinel plants  
318 close to the sown flower patches as in those close to the resident vegetation. However,  
319 for the other natural enemies' groups, the proximity of the sown flower patches did not  
320 change their abundance. These results could be influenced by the orchards' sizes or by  
321 the proximity of both groups of sentinel plants (close to the sown flower patches vs.  
322 close to the resident vegetation), which ranged from 25 to 130 m, compared to the nat-  
323 ural enemies' flight capacity. It has been suggested that parasitoids and some predators,  
324 including anthocorids and coccinellids, can move long distances between crops, mainly  
325 during the spring (Pons and Stary 2003, Lumbierres et al. 2007). It has also been de-  
326 scribed that hoverflies can fly distances of up to 200 m (Wratten et al. 2003), and van  
327 Schelt and Mulder (2000) found *A. aphidimyza* eggs on plants at distances of up to 45 m  
328 from the release point.

329 *Aphidoletes aphidimyza* was recorded as the most abundant predator in the sentinel  
330 plants, but it was never found in either the insectary plants or in the resident vegetation.  
331 Although it has been described as being attracted by flowers, such as *L. maritima* (Apa-  
332 ricio et al. 2018), the absence of individuals obtained with the beating was probably due  
333 to their behavior and to the fact that the beating was conducted on the upper parts of



334 the plants. These adults only fly at dusk and during the night (Harris 1973), and in day-  
335 time, they hide in the shaded parts of the plant (Boulanger et al. 2019). Because the  
336 weather during the light hours in the study area is hot and dry, it is then conceivable  
337 that adults remain immobile in the lower parts of the vegetation where temperatures  
338 are milder and the humidity is higher.

339 According to the obtained results, hymenopteran parasitoids and hoverflies were more  
340 attracted to the sown flower patches than to the resident vegetation, probably due to  
341 the more abundant and prolonged flower presence in the insectary plant patches, which  
342 turned out in provision of more suitable and permanent food resources. The abundance  
343 of flowers might have also affected the relative abundance of natural enemies on the  
344 different insectary plants (Ambrosino et al. 2006). Feeding on flower nectar is common  
345 in hoverflies and hymenoptera parasitoids, which use this sugar-rich resource mainly to  
346 fuel their foraging for food resources, mating and oviposition sites (Wäckers 2005, Nic-  
347 olson and Thornburg 2007). Additionally, floral resources such as nectar and pollen are  
348 used by hoverflies as a protein source for ovary maturation and egg production (Bran-  
349 quart and Hemptinne 2000, Van Rijn et al. 2013). Similarly, some flowers have shown to  
350 have a positive effect on the reproduction of aphid parasitoids (Araj and Wratten 2015,  
351 Aparicio et al. 2018).

352 In the present samplings, *L. maritima* was an attractive resource for hoverflies and par-  
353 asitoids. Our results agree with those of previous studies reporting that the addition of  
354 *L. maritima* in the field improved the abundance of hoverflies (Hogg et al. 2011, Gontijo  
355 et al. 2013) and hymenopteran parasitoids (Sivinski et al. 2006, Rohrig et al. 2008). Ad-  
356 ditionally, under laboratory conditions, it has been shown that the availability of this

357 floral resource enhances the fitness of some parasitoid species (Arnó et al. 2018, Apari-  
358 cio et al. 2018, Johanowicz and Mitchell 2000). *Achillea millefolium* attracted hymenop-  
359 tera parasitoids but not hoverflies. The attraction to blooming *A. millefolium* has been  
360 reported for parasitoids (Dib et al. 2012, Lundin et al. 2019) and for hoverflies (Colley  
361 and Luna 2000). During the present samplings, *A. millefolium* bloomed only from the  
362 end of May 2016 onward. Despite this lack of blooming, the plant was highly attractive  
363 to adult wasps, suggesting that compounds other than those coming from flowers are  
364 involved in the attractiveness to these natural enemies. *Sinapis alba* and *M. arvensis*  
365 were attractive for hoverflies but did not attract other natural enemies. However, it has  
366 been previously described that *S. alba* is beneficial for some parasitoid species (Vattala  
367 et al. 2006, Arnó et al. 2018) and for Syrphidae (Carreck 1997).

368 The presence of ladybeetles during the first year of sampling was higher in the insectary  
369 plants than in the resident vegetation, especially in *L. maritima* and *S. alba*, and although  
370 without significant differences, the same pattern was observed for *Orius* spp in *L. mari-  
371 tima*. Being omnivores, both predators benefit from feeding on nectar and pollen, and  
372 they use plants' nonflowering parts to rest, mate, egg lay, complete preimaginal devel-  
373 opment, and take shelter (Landis et al. 2000, Coll and Guershon 2002). Therefore, the  
374 addition of insectary plants can enhance these predators' presence in crops and contrib-  
375 ute to the biological control of pests such as aphids and thrips.

376 The aphid species present in the insectary plants or the resident vegetation were not  
377 identified as *M. persicae* or *Hyalopterus* spp. Therefore, our results suggested that these  
378 insectary plants are not reservoirs of damaging aphids for peach crops in the study area.  
379 Concerning thrips, several morphospecies were detected during the samplings, some of

380 which resembled *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), an im-  
381 portant pest in nectarine cultivars (Gonzalez et al. 1994, Avilla et al. 2008). Although the  
382 presence of these herbivores could be considered a negative trait for insectary plants  
383 near crops, they might also play an important role as alternative prey in establishing and  
384 expanding natural enemy populations in insectary plants (Norris and Kogan 2000, Boivin  
385 et al. 2012).

386 In summary, there are several predators associated with *M. persicae* in peach orchards,  
387 *A. aphidimyza* and *E. balteatus* being the most abundant. These two species together  
388 with the parasitoid *A. matricariae* (Aparicio et al. 2019) must be considered the key bi-  
389 ological control agents of this aphid in peach orchards. In our study, the guild of natural  
390 enemies was the same on both the sown flower species and the resident vegetation.  
391 However, hoverflies clearly preferred sown flower patches and, parasitoids preferred *L.*  
392 *maritima* and *A. millefolium* to the resident vegetation. Although in our experimental  
393 set up the four insectary plants were placed together and attraction to one of the spe-  
394 cies might mask lack of attraction of another, *L. maritima* and *A. millefolium* should be  
395 considered as key candidates in the design of successful ecological infrastructures. In  
396 addition to the attractiveness for natural enemies, several other aspects have to be  
397 taken into account when designing the ecological infrastructures: first, the capability of  
398 selected candidate plants to effectively supply resources to the natural enemies (Wäck-  
399 ers 2005); second, ensure a minimum size of the surface with flowers in relation to the  
400 size of the field (Blaauw and Isaacs 2012); and finally, the adaptation of the selected  
401 plants to the environmental and agronomic conditions. Additionally, further studies are  
402 needed to verify the contribution of the establishment of floral field margins to the bio-  
403 logical control of aphids and other peach pests.

404

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413

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591

592 **TABLES**

593 **Table 1.** Description of the phenological stages for each plant species during the sam-  
 594 pling periods of 2015 and 2016: in bloom (✿), vegetative (🌿), and dry (▪).

Insectary plants	Phenological stage							
	2015				2016			
	Week				Week			
	18	20	22	24	18	20	22	24
<i>L. maritima</i>	✿	✿	✿	✿	✿	✿	✿	✿
<i>M. arvensis</i>	✿	✿	✿	✿	✿	✿	✿	✿
<i>S. alba</i>	✿	✿	✿	✿	✿	✿	▪	▪
<i>A. millefolium</i>	🌿	🌿	🌿	🌿	🌿	🌿	✿	✿

595

596 **Table 2.** Dates in which the different plant genera were in bloom and present (✿) in  
 597 more than 25 % of the surfaces of the resident vegetation patches.

Plant genera	2015			2016		
	20	22	24	20	22	24
<i>Galium</i>	✿	✿		✿	✿	✿
<i>Malva</i>	✿	✿	✿	✿	✿	✿
<i>Hordeum</i>	✿					✿
<i>Trifolium</i>		✿				
<i>Avena</i>		✿	✿			
<i>Bromus</i>		✿	✿			
<i>Carduus</i>				✿	✿	✿
<i>Sonchus</i>					✿	
<i>Convolvulus</i>						✿

598

599 **Table 3.** Total numbers of natural enemies collected in the sentinel plants in 2015/2016  
 600 sampling seasons

Field	Total parasitoids	Hoverflies	Cecidomids	Anthocorids	Coccinelids	Lacewings	Total predators
1	69 / 134	36 / 7	10 / 15	13 / 0	0 / 2	4 / 0	63 / 24
2	51 / 185	10 / 5	3 / 13	10 / 0	2 / 5	1 / 3	26 / 26
3	110 / 59	25 / 6	78 / 29	4 / 0	2 / 3	3 / 3	112 / 41
4	208 / 76	12 / 1	37 / 37	5 / 0	0 / 3	0 / 2	54 / 43

601

602

603

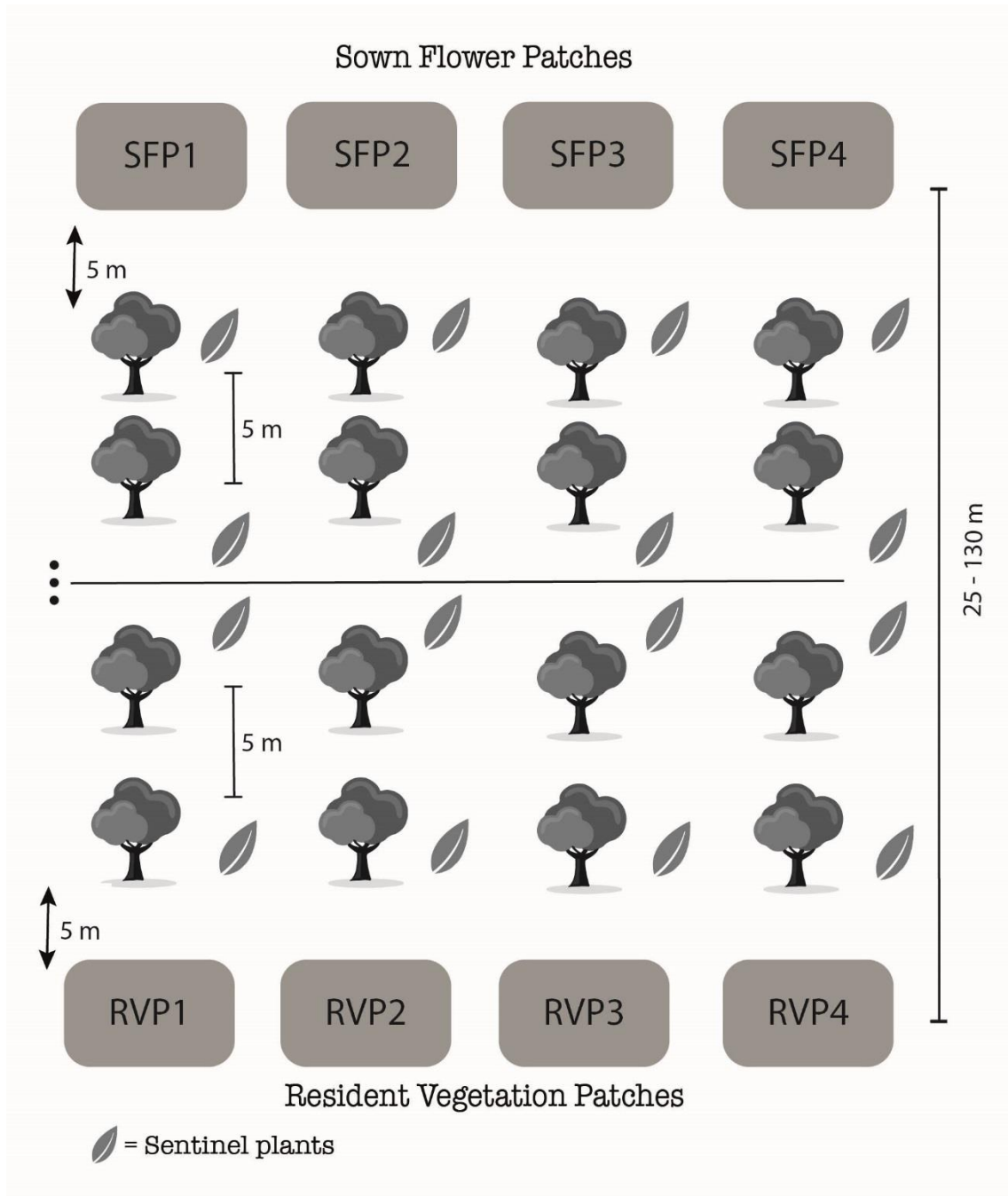
604 **Table 4.** Natural enemies of the *M. persicae* (mean  $\pm$  SE) recorded with sentinel plants close to the sown flower patches and to the resident  
 605 vegetation in each sampling year.

Year	2015				2016			
	Near sown flower patches	Near resident vegetation	Z	P	Near sown flower patches	Near resident vegetation	Z	P
Parasitoids	1.54 $\pm$ 0.329	1.92 $\pm$ 0.278	-1.225	0.220	2.18 $\pm$ 0.391	3.13 $\pm$ 0.716	-0.916	0.360
Hoverflies	0.43 $\pm$ 0.087	0.22 $\pm$ 0.087	1.996	<b>0.046</b>	0.11 $\pm$ 0.045	0.10 $\pm$ 0.033	0.274	0.784
Cecidomids	0.41 $\pm$ 0.161	0.59 $\pm$ 0.162	-1.161	0.246	0.62 $\pm$ 0.340	0.45 $\pm$ 0.178	0.527	0.593
Anthocorids	0.11 $\pm$ 0.040	0.13 $\pm$ 0.038	-0.302	0.763				
Coccinellids					0.05 $\pm$ 0.023	0.10 $\pm$ 0.042	-0.974	0.333

606

607

608 **FIGURE LEGENDS**

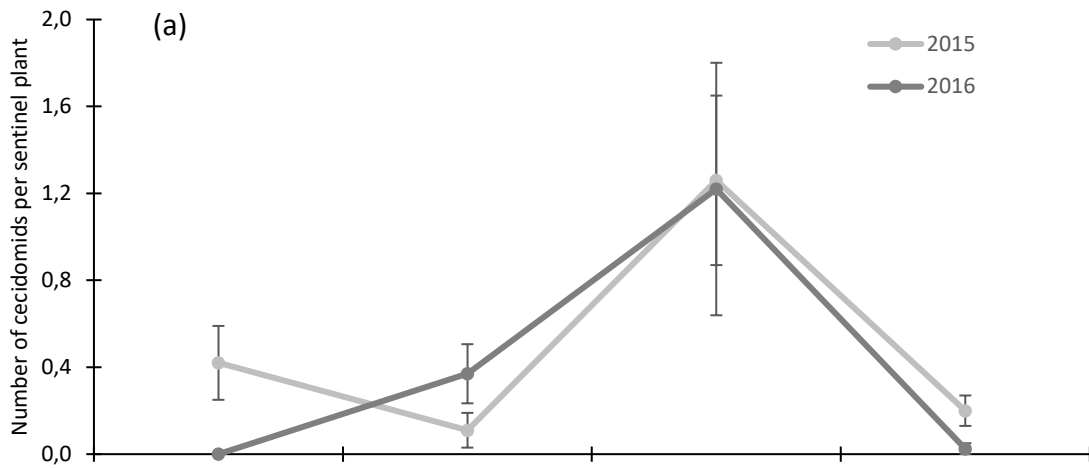


609

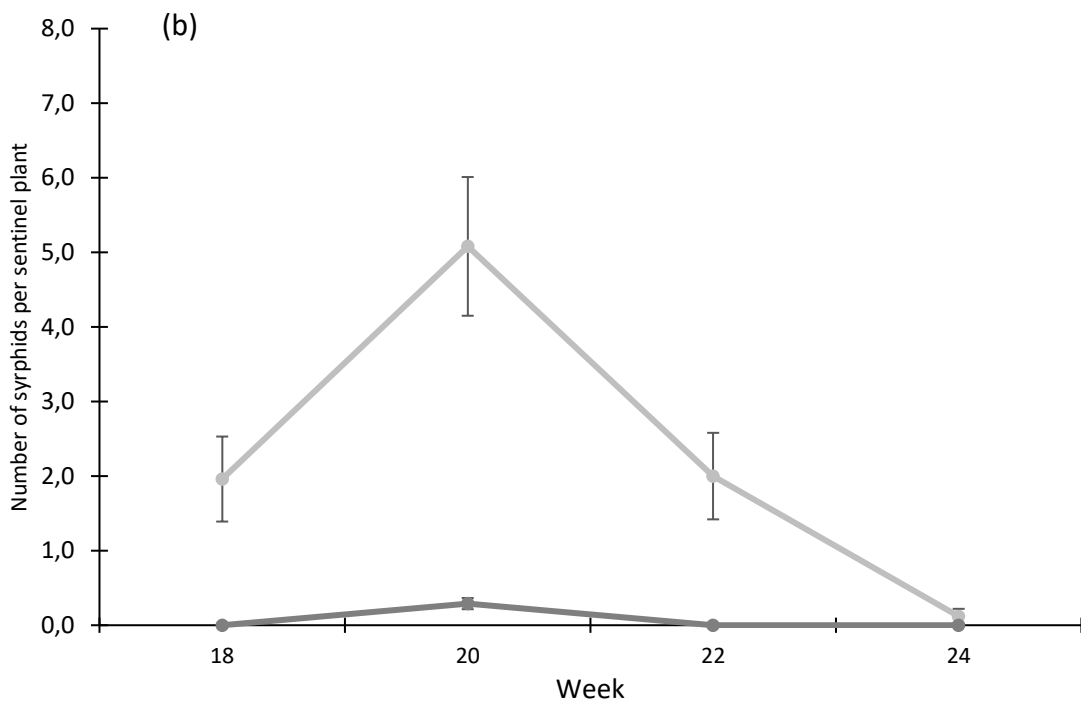
610 **Fig. 1.** Experimental set-up in each of the four peach orchards.

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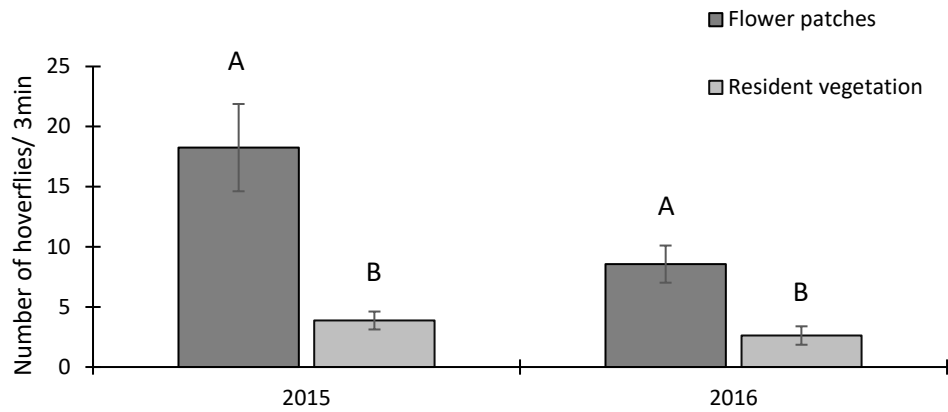
616 **Fig. 2.** Seasonal abundances of Cecidomyiidae (a) and Syrphidae (b) larvae (mean ± SE)

617 recorded in the peach sentinel plants placed in the peach orchards.

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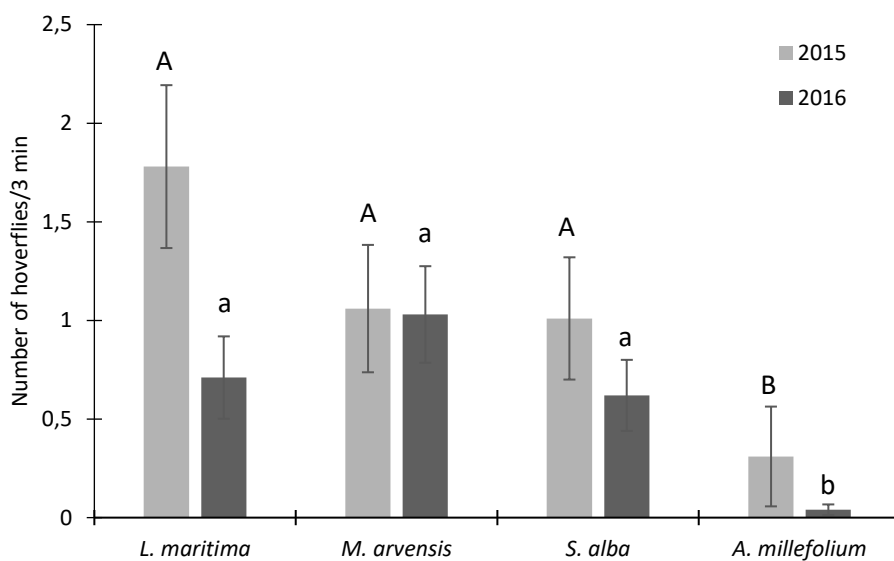




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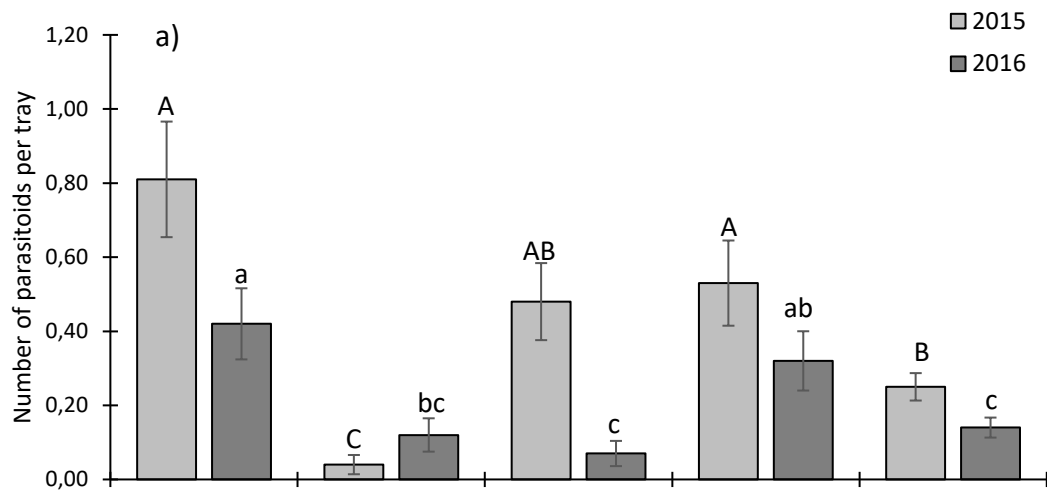
621 **Fig. 3.** Number of adult hoverflies (mean  $\pm$  SE) hovering above the sown flower and res-  
 622 ident vegetation patches for three minutes in each sampling year. For each year, differ-  
 623 ent uppercase letters indicate significant differences among the patches ( $P < 0.05$ ).

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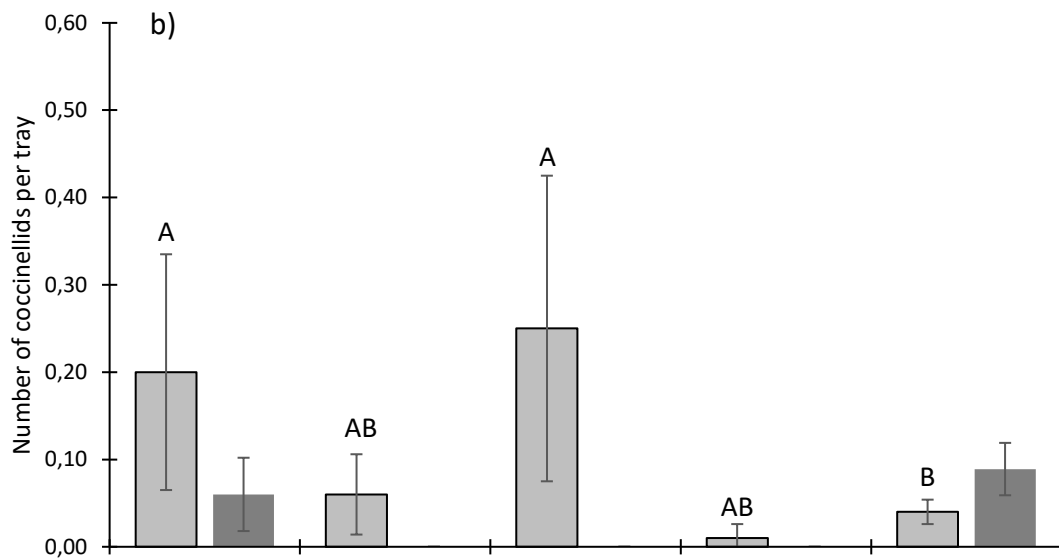


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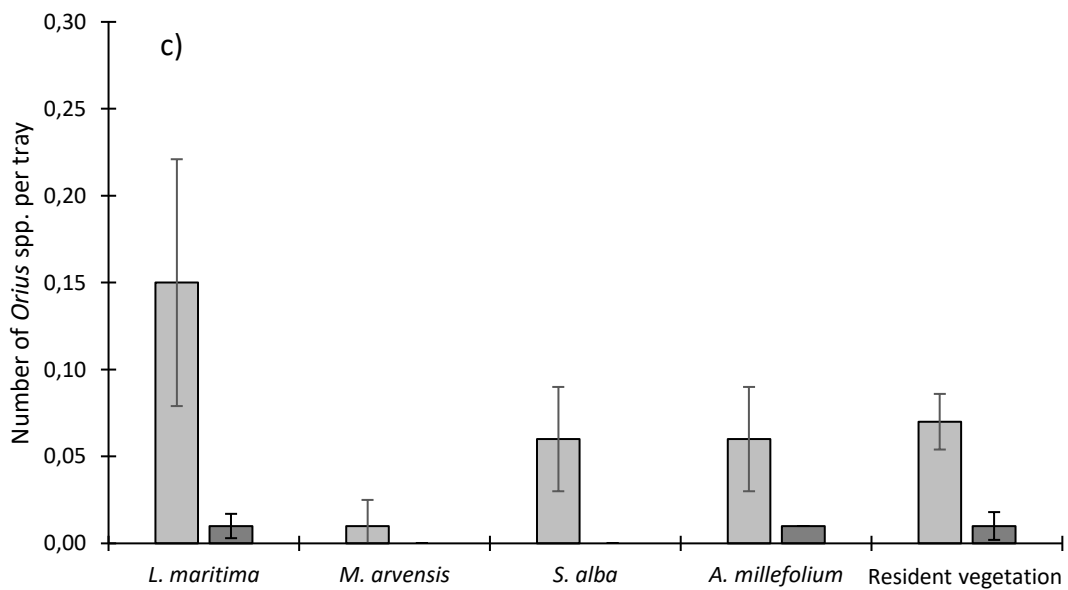
626 **Fig. 4.** Number of hoverfly landings (mean  $\pm$  SE) per insectary plant during the three  
 627 minutes' observation of sown flower patches in both sampling years. The different let-  
 628 ters indicate significant differences among the insectary plants in 2015 (upper case) and  
 629 in 2016 (lower case) ( $P < 0.05$ ).



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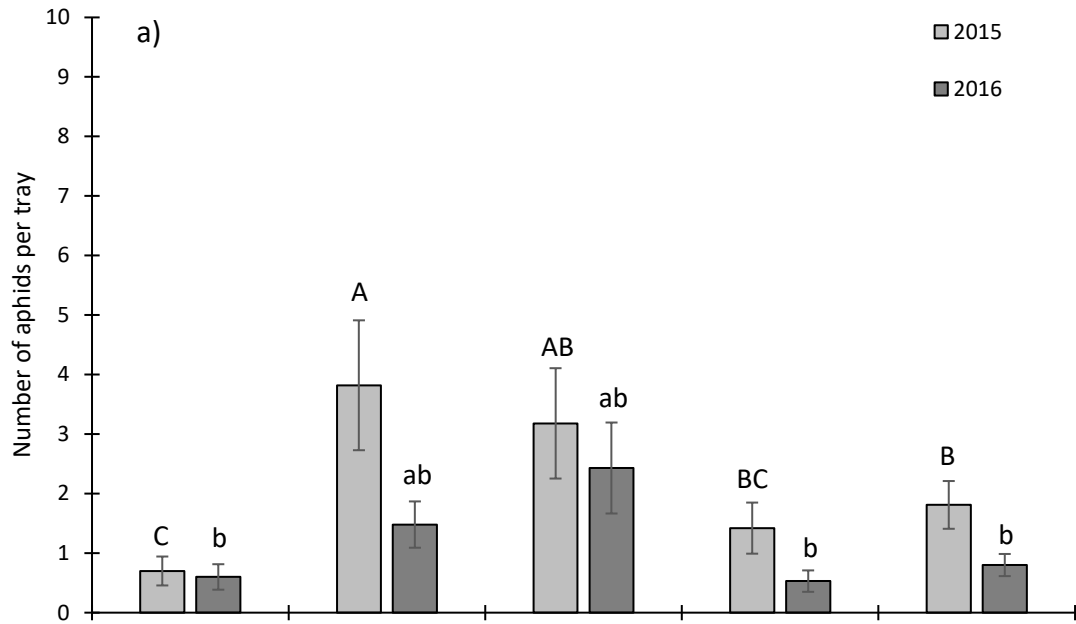


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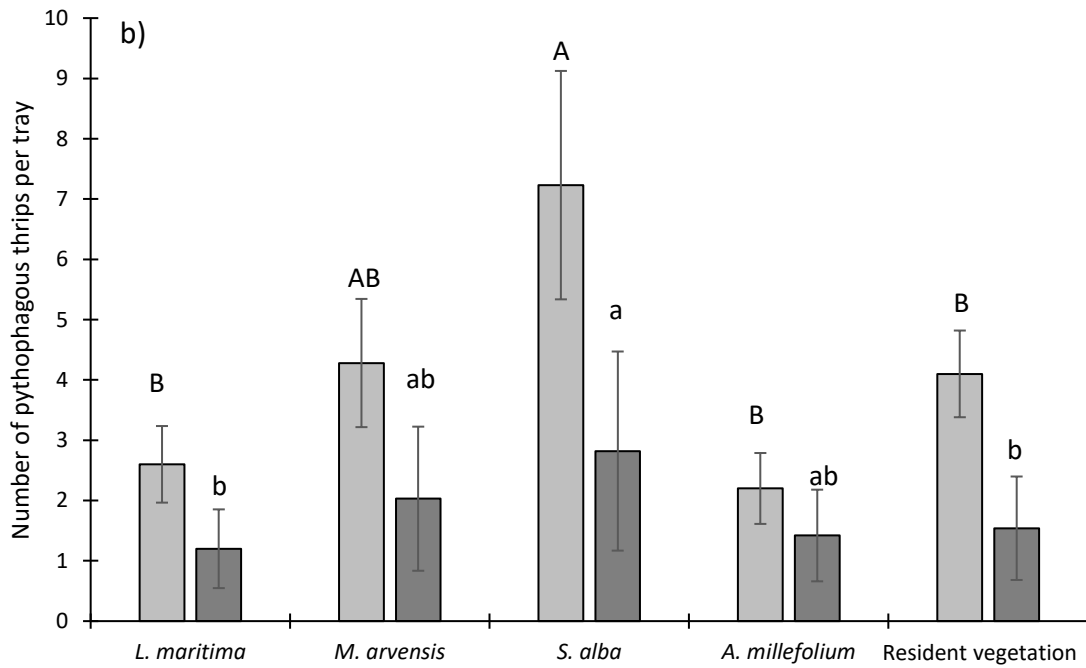
633 **Fig. 5.** Number of hymenopteran parasitoids (a), Coccinellidae (b) and *Orius* spp. (c)  
634 (mean  $\pm$  SE) recorded per insectary plant and per resident vegetation via beating during  
635 both sampling years. The different letters indicate significant differences among the in-  
636 sectary plants in 2015 (upper case) and in 2016 (lower case) ( $P < 0.05$ ).

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641 **Fig. 6.** Number of aphids (a) and thrips (b) (mean ± SE) recorded per insectary plant and  
 642 per resident vegetation via beating during both sampling years. The different letters in-  
 643 dicate significant differences among the insectary plants in 2015 (upper case) and in  
 644 2016 (lower case) ( $P < 0.05$ ).