

**Assessing the relationship between pest density and plant damage: a case study with the belowground herbivore *Delia radicum* (Diptera: Anthomyiidae) on broccoli**

Xavier Mesmin, Marie Vincent, Yann Tricault, Vianney Estorgues, Loic Daniel, Marie Cortesero, Vincent Faloya, Anne Le Ralec

► **To cite this version:**

Xavier Mesmin, Marie Vincent, Yann Tricault, Vianney Estorgues, Loic Daniel, et al.. Assessing the relationship between pest density and plant damage: a case study with the belowground herbivore *Delia radicum* (Diptera: Anthomyiidae) on broccoli. *Applied Entomology and Zoology*, Springer Verlag, 2019, 54 (2), pp.155-165. 10.1007/s13355-019-00607-3 . hal-02063365

**HAL Id: hal-02063365**

**<https://hal-agrocampus-ouest.archives-ouvertes.fr/hal-02063365>**

Submitted on 22 Mar 2019

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1 **Assessing the relationship between pest density and plant damage: a case study with**  
2 **the belowground herbivore *Delia radicum* (Diptera: Anthomyiidae) on broccoli**

3  
4 Xavier Mesmin <sup>a</sup>, Marie Vincent <sup>a</sup>, Yann Tricault <sup>b</sup>, Vianney Estorgues <sup>c</sup>, Loïc Daniel <sup>d</sup>, Anne-  
5 Marie Cortesero <sup>a</sup>, Vincent Faloya <sup>d</sup>, Anne Le Ralec <sup>a</sup>

6  
7 <sup>a</sup> IGEPP, Agrocampus Ouest, INRA, Université de Rennes 1, Université Bretagne-Loire, 35000  
8 Rennes, France

9 <sup>b</sup> IGEPP, Agrocampus Ouest, INRA, Université de Rennes 1, Université Bretagne-Loire, 49000  
10 Angers, France

11 <sup>c</sup> Chambre régionale d'agriculture de Bretagne, Kergompez, 29250 Saint-Pol-de-Léon, France

12 <sup>d</sup> IGEPP, Agrocampus Ouest, INRA, Université de Rennes 1, 35650 Le Rheu, France

13  
14 **Abstract**

15 For many crops, we have poor knowledge about the relationship between pest density  
16 and damage. However, investigating pest harmfulness is particularly relevant currently in the  
17 search for alternative crop protection strategies that are unlikely to totally suppress pest  
18 populations. Here, we assessed the harmfulness of *Delia radicum* (L.) on broccoli (*Brassica*  
19 *oleracea* var. *italica* Plenck). We worked inside insect-proof cages set up in the field with  
20 additional pitfall traps to remove ground dwelling predators. Plants were manually infested with  
21 10 levels of pest density ranging from 0 to 100 individuals per plant, following a natural  
22 infestation pattern. Surprisingly, no plants died but almost 100% of the pests introduced died  
23 over the course of the experiment. However, all broccoli development and growth traits were  
24 negatively correlated with pest density and broccoli head mass at harvest decreased linearly  
25 with pest density. The observation over time of development and growth traits showed evidence  
26 of plant compensation, suggesting that the head mass of individual plants may have reached  
27 similar values if allowed to fully mature. The relationship between pest density and damage,  
28 together with forecast models of pest population dynamics could be used to develop decision  
29 support tools assessing the relevance of preventative treatments.

30  
31 **Keywords**

32 Crop losses; plant traits; plant injury; damage; plant compensation.

33  
34 **Introduction**

35 Throughout the history of agriculture, the development of pest management strategies  
36 has led to dramatic decreases in crop losses to pests (Oerke 2006). However, the predominant  
37 crop protection method, that is pesticide usage, has various negative consequences, notably on  
38 human health and the beneficial fauna of agro-ecosystems (Desneux et al. 2007; Geiger et al.  
39 2010). Alternatives are therefore urgently needed, but in contrast to pesticides, most of these  
40 alternatives (push-pull, insect netting, intercropping...) are likely to have partial efficiency. For  
41 example, conservation biological control, based on the enhancement of natural enemy  
42 populations, is thought to be an interesting method for reducing crop losses (Eilenberg et al.  
43 2001) but is incompatible with the complete suppression of pest populations because by

44 definition, predators cannot maintain their populations without prey. It is therefore necessary to  
45 reconsider the relationship between pest density, plant injuries (i.e. the symptoms of pest  
46 development on the physiology of the host; Pedigo et al. 1986) and damage (i.e. the decrease  
47 in yield quantity or quality; Zadoks 1985; Pedigo et al. 1986). In this context, pest populations  
48 should reach an intermediate size that is both i/ high enough to allow the build up of efficient  
49 natural enemy communities, including several specialists depending on this specific resource  
50 (Devictor et al. 2010) and ii/ low enough to result in acceptable losses for the farmer, a level  
51 that is likely to depend on the production situation (*sensu* Aubertot and Robin 2013)  
52 (International Conference on Global Crop Losses 2017). In many cases, plants are able to  
53 tolerate moderate levels of pest infestation (Poston et al. 1983; Fenemore 1984; Pedigo et al.  
54 1986), i.e. to withstand injuries without significant damage (Verdugo et al. 2016), so that this  
55 intermediate pest population size could be innocuous in several pest/crop systems (e.g. Rogers  
56 and Brier 2010).

57 In general, injuries and damage due to below ground pests have been less well studied  
58 than those inflicted by above ground pests (Hunter 2001). This may be due to methodological  
59 constraints because recording freshly caused injuries below ground necessarily involves  
60 destructive sampling, thus preventing the evaluation of subsequent damage. However, the  
61 effects of pest development below ground are not restricted to root tissue loss and indirect  
62 effects of root injuries could also occur above ground (Teixeira et al. 1996; Murray et al. 1996;  
63 Hunter 2001). Indeed, the physiological and morphological consequences of root injuries can  
64 include reduced leaf surface (Cardona et al. 1982), plant height and above ground biomass  
65 (Boica Junior et al. 2015), as well as an increase in lignin content (Hopkins et al. 1995), and  
66 soluble nitrogen concentration in the sap (Gange and Brown 1989). Gange and Brown (1989)  
67 suggested that due to root removal, below ground pests could have a similar effect as drought  
68 on plants. These indirect effects which can be assessed by monitoring plant growth and  
69 development above ground, may or may not lead to a significant decrease in final yield  
70 (Rosenheim et al. 1997; Brandelero et al. 2016).

71 *Delia radicum* (L.) (Diptera: Anthomyiidae) is the main pest of *Brassica* vegetables in  
72 northwestern Europe. The main cause of damage due to this pest in leaf and flower vegetables  
73 is early plant mortality, which can reach 40-60% without insecticides (Estorgues 2005).  
74 Females lay their eggs on the ground, within a distance of 5cm from plant stems (Hughes and  
75 Salter 1959). The larvae then develop by feeding on plant roots throughout three life stages.  
76 They thereby inflict injuries to the roots with several potential consequences on the plants (see  
77 above) among which the disturbance of water and nutrient uptake (Gange and Brown 1989).  
78 Several studies have been performed to quantify the damage inflicted by this pest on two  
79 *Brassica* crops: cauliflower and oilseed rape. Turnock et al. (1992) did not find a relationship  
80 between injuries caused by root maggots and yield of oilseed rape, hence suggesting that  
81 cropped plants fully compensated for injuries, at least up to the maximal tested pest density (25  
82 eggs per plant). In cauliflower, El Titi (1979) showed a correlation between egg density and  
83 plant mortality for infestation levels ranging from 10 to 60 eggs per plant. This study was used  
84 by Bligaard et al. (1999) to define an economic threshold of *D. radicum* on cauliflower,  
85 suggesting that intervention was needed if more than one egg was found per felt trap (used for  
86 monitoring egg laying) and per day two to four weeks after planting. This threshold is still used  
87 by farmers and advisors in France to evaluate the intensity of pest infestation, but it is no longer

88 relevant in terms of decision making because: i/ the chlorfenvinphos used by El Titi (1979) is  
89 now prohibited and ii/ the current protection strategy against the cabbage root fly is preventative  
90 and consists of drenching the roots of all leaf and flower *Brassica* vegetables with spinosad  
91 (Success™ 4) before planting. Also in cauliflower, Bligaard (1999) performed several  
92 experiments with varying times of infestation and found that plant biomass decreased when egg  
93 density rose from 0 to 25 eggs per plant, but final yield was not monitored. Only one study  
94 showed reduced production of surviving cauliflowers in response to *D. radicum* attack (El Titi  
95 1977). Finally, a methodological limitation in these previous studies was that the eggs were  
96 always introduced all at once. This pattern is unrealistic compared to natural infestation  
97 (Estorgues 2005) and may have had consequences on pest survival, plant-pest interactions and  
98 therefore on the subsequent damage: Finch and Skinner (1988) showed increased *D. radicum*  
99 survival for instant egg inoculation compared to protracted inoculation.

100 The objective of the present study was to assess *D. radicum* harmfulness on broccoli  
101 *Brassica oleracea* var. *italica* Plenck (Brassicales: Brassicaceae), a crop for which we expected  
102 a similar tolerance as that previously observed for cauliflower. For this, we worked on caged  
103 plants grown in the field to ensure natural root development and we controlled pest density  
104 using artificial egg infestations. We expected broccoli to show a tolerant response to *D. radicum*  
105 (Poston et al. 1983), with compensation for low injury levels, hence no damage; and damage  
106 resulting from greater levels of injury, ranging from yield reduction to plant mortality.

## 107 **Materials and methods**

### 108 *Study site and experimental design*

109  
110  
111 The experiment was conducted in an 850m<sup>2</sup> plot at INRA's experimental station  
112 (UE0787, Domaine expérimental de la Motte au Vicomte), in Le Rheu, France (48°06'N;  
113 1°47'W) during spring 2016. The field used for this study has a deep soil (80-120cm),  
114 hydromorphic from the surface, originating from shale and wind deposited loam (classified as  
115 luvisol following the international soil classification system of the IUSS Working group WRB  
116 2015). Previous crops were maize in 2012, 2013, 2014; fava beans in 2015 and meadow for  
117 seven months before this study started. Six insect-proof cages (6m long × 3m wide × 2m high;  
118 300 \* 300µm mesh; Diatex ®) were set up on the plot. On the 5<sup>th</sup> of April 2016, 30 untreated  
119 broccoli plants (cv. 'Marathon') at the stage of two true leaves were planted in each cage, every  
120 0.50m in rows 0.75m apart. Insect-proof cages prevented plant colonization by flying insects,  
121 whether they were females from surrounding cabbage root fly populations, other *Brassica* pests  
122 or *D. radicum* natural enemies. PET barriers (Greenborder, Nortene®) buried all around the  
123 cages (60cm high, 40 of which below ground) prevented hypogeic and epigeic organisms from  
124 gaining access. Two sticky panels (29.7 \* 42cm), a yellow and a blue one, were hanged inside  
125 each cage in order to catch flying arthropods that could emerge from the soil. Twenty four  
126 pitfall traps were also set up in each cage (i.e. 1.3 per m<sup>2</sup>) to reduce predation by ground  
127 dwelling arthropods on immature fly stages. Pitfall traps were half-filled with water and a few  
128 drops of odorless detergent. Barriers and traps were set up 13 days before planting, and the  
129 cages, the day before planting.

131 *Artificial plant infestation*

132

133 Eggs were manually deposited on plants inside insect-proof cages. The timing of egg additions  
134 was designed to mimic the natural egg laying dynamics of *D. radicum*, which is generally  
135 centered on a peak that represents about 40% of the total amount laid (calculation based on  
136 previous experiments performed under natural infestation conditions). The first natural egg  
137 laying peak is quite stable over the years in north-western France and typically occurs between  
138 early and mid-May when transplanting is done at the beginning of April (Estorgues 2005). We  
139 hence spread the infestation out over three consecutive weeks with a peak on the 2<sup>nd</sup> of May  
140 representing 40% of the total amount of eggs applied on each plant. The two other inputs were  
141 made on the 25<sup>th</sup> of April and on the 9<sup>th</sup> of May, each representing 30% of the total amount of  
142 eggs applied per plant. We defined ten pest densities, summarized in Table 1, ranging from 0  
143 (control) to 100 eggs per plant, a range similar to egg counts made in production fields from  
144 the same region (Josso et al. 2013).

145 The eggs were obtained in the laboratory from a strain of *D. radicum* originally collected  
146 in fields of the same experimental station in early summer 2015 and reared as described in  
147 Lamy et al. (2017). Females were offered slices of swede placed on a filter paper for egg laying.  
148 The eggs were then brought to the field and directly used for infesting the plants. They were  
149 deposited at the base of plant stalks with a fine paintbrush. At each date of plant infestation, an  
150 additional batch of about 300 eggs was placed in a Petri dish, on a moistened filter paper and  
151 kept in a climate-controlled room (16:8h photoperiod and 20°C). After seven days, natural egg  
152 mortality was estimated as the proportion of unhatched (i.e. dead) eggs. Infestation levels were  
153 distributed to broccoli plants in a randomized complete block design: inside each cage, egg  
154 density was attributed randomly to each plant; there were three broccoli plants for each egg  
155 density in each cage, thus a total of 18 plants per egg density in the experiment.

156 At broccoli harvest, i.e. on June 21 and 22, a soil sample (12cm in diameter and  $13.5 \pm$   
157  $0.4$ cm (mean  $\pm$  SE) in depth, ensuring the collection of more than 70% of the pupae; Hughes  
158 1960; Finch et al. 1978) was taken around each broccoli root with a motorized auger. The  
159 number of *D. radicum* larvae and pupae was counted after washing the samples through a 1mm  
160 \* 1mm mesh sieve.

161

162 *Plant development and growth*

163

164 We evaluated plant development and plant growth weekly for every broccoli plant from  
165 April 12 to June 14 (i.e. during 10 weeks). The development of each plant was assessed by  
166 recording the leaf numbers, the presence / absence of at least one lateral sprout and the presence  
167 / absence of a visible inflorescence. These measurements match the three main stages of  
168 broccoli development (Feller et al. 1995). Plant mortality was also recorded. Plant growth was  
169 evaluated via two measurements: the product of the largest leaf length and width (thereafter  
170 called “relative leaf area”), known to be highly correlated with leaf area in *Brassica* crops  
171 (Olfati et al. 2010; Cargnelutti Filho et al. 2015; Tartaglia et al. 2016) and the height above  
172 ground of the apical meristem (thereafter simply called “plant height” in the text), often used to  
173 give complementary information on plant growth (Kloen and Altieri 1990; Brandelero et al.  
174 2016).

175

176 *Root injuries and final head mass*

177

178 As the first broccoli had reached their optimal development regarding marketable  
179 standards (buds tight and close to opening), we harvested every plant in a row on the 21<sup>st</sup> and  
180 22<sup>nd</sup> of June. Stalks were cut about 1cm below the insertion of the first branch of the head and  
181 heads were weighed with a spring scale (precision =  $\pm 5$ g).

182 Injuries caused by *D. radicum* larvae feeding on plant roots was assessed by visual  
183 examination using the scale defined by Dodsall et al. (1994): 0 = no injury; 1 = slight feeding,  
184 < 10% of tap root surface injured; 2, 3, 4 and 5 respectively 10-25%, 26-50%, 51-75%, 76-  
185 100% of root surface injured.

186

187 *Data analysis*

188

189 Generalized linear mixed models were used to analyze every dimension of plant  
190 development, growth and production. We generated a model for every dimension of plant  
191 development (i.e. number of leaves, proportion of plants with at least one lateral shoot and  
192 proportion of plants with an inflorescence) and growth (i.e. relative largest leaf area, plant  
193 height) including the number of eggs (quantitative), the sampling session (factor), and their  
194 interaction as fixed effects as well as the cage and plant identifiers as random effects to account  
195 respectively for potential spatial correlation among data obtained inside the same cage and for  
196 the fact that the same plants were monitored in each session (Faraway 2006). The analysis of  
197 lateral shoots and inflorescences was restricted to the dates when shoots or inflorescences were  
198 recorded, respectively. For the latter, only two measurement sessions were analyzable which  
199 made it impossible to use the plant identifier as a random factor. This proportion was therefore  
200 analyzed at the cage scale and the random effect set for the cage identifier to account for  
201 repeated measures (Faraway, 2006). Pearson correlation coefficients were computed among all  
202 development and growth traits over their respective period of analysis (Table S1 in  
203 Supplementary materials).

204 The effect of the number of eggs on the proportion of plants displaying root injuries  
205 (injury class > 0) and on final head mass was assessed, using a random factor set for the cage  
206 identifier to consider potential spatial correlation among data obtained inside the same cage.

207 All statistical analyses were performed using R software (R core team 2017). The  
208 models described above were fitted using generalized linear mixed modeling (functions ‘lmer’  
209 or ‘glmer’ of the package ‘lme4’; Bates et al. 2015) with a distribution and link function adapted  
210 to the data analyzed: identity-link Gaussian (response variables: log transformed number of  
211 leaves; log transformed relative largest leaf area; square root transformed plant height; final  
212 head mass) or logit-link binomial (response variables: proportion of plants with at least one  
213 lateral shoot; proportion of plants with an inflorescence; proportion of plants with root injuries).  
214 When necessary, an additional random factor set for the statistical individual was used to  
215 account for data overdispersion. The significance of the fixed effects was tested using type II  
216 Wald chi-square tests (function ‘Anova’, package ‘car’; Fox and Weisberg 2011).

217 Post-hoc tests were performed on the models fitted on plant development and growth  
218 traits. We studied the effect of egg density on plant traits as a function of time: for each date

219 (factor level), we estimated the marginal slope of the linear trend between egg density and plant  
220 traits (in cm/egg for plant height for instance; function ‘*emtrends*’ with back-transformation of  
221 the slopes to the response scale; package ‘*emmeans*’; Lenth 2017). Then we extracted the slope  
222 of the steepest relationship between each plant trait and egg amount. These values provided  
223 estimates for the strongest effects of pests on plant traits observed in our experimental design.

224 Thirty plants (i.e. 17%) suffered from slug attack or lost their apical meristem so that it  
225 was not possible to record their largest leaf length and width or plant height on at least one  
226 occasion. All data collected on such plants were discarded.

## 227 **Results**

228

### 229 *Plant survival and mortality of D. radicum immature stages*

230

231 The 150 broccoli plants survived the entire experiment. The proportion of unviable eggs  
232 among the three batches used for artificial infestations was similar with an overall mean of  
233 18.9% ( $\pm 1.3\%$ ). At the end of the experiment, a total of only two pupae was recovered from  
234 the soil samples taken around the plant roots, suggesting a high mortality of the 6065 introduced  
235 eggs before reaching their final developmental stage.

236

### 237 *Plant development and growth*

238

239 The relative largest leaf area, plant height and the number of leaves were highly  
240 correlated, with Pearson correlation coefficients exceeding 90%. In contrast, none of the data  
241 had a correlation coefficient exceeding 63% with the proportion of plants with an inflorescence  
242 and coefficients did not exceed 37% for correlations with the proportion of plants with at least  
243 one lateral shoot (Table S1 in Supplementary materials).

244 Except for the proportion of plants with at least one lateral shoot, all development and  
245 growth traits were negatively correlated with an increase in pest density (Table 2). The  
246 interaction between measurement dates and the number of eggs deposited was significant for  
247 growth traits, but not significant for development traits.

248 Plant trait responses to pest attack followed a similar pattern (Fig. 1) of 1) no pre-  
249 existing trend in development and growth traits prior to pest input; then 2) a transient negative  
250 effect becoming significant after a delay varying from a couple of days to several weeks after  
251 the first infestation, depending on the trait, and finally 3) a waning of the effect with no  
252 significant trend at the end of the experiment. Hence, even the highly infested plants recovered  
253 and showed the same final development and growth states as those of non-infested plants, as  
254 measured by our parameters. The exception was plant height, which remained negatively  
255 correlated with egg density until the end of the experiment. All plants produced an inflorescence  
256 and 73% produced at least one lateral shoot by the end of the experiment. The time course of  
257 negative effects of egg density (Fig. 1) indicates that these organs appeared later when plants  
258 suffered high pest densities.

259 The date at which the most negative trend between egg density and development or  
260 growth trait was observed varied depending on the trait considered. It ranged from 23 days to  
261 50 days after the first artificial egg infestation for the proportion of plants with lateral shoots  
262 and the plant height respectively (Fig. 1). Focusing on this particular date, the models indicated

263 linear relationships between plant traits and egg density (Fig. 2), with only slight deviations  
264 from linearity for the proportion of plants with at least one lateral shoot or with an inflorescence,  
265 which may simply be due to the binary nature of these response variables.

266

#### 267 *Root injuries and final head mass*

268

269 All plants harvested were in injury class 0 or 1 (i.e. less than 10% of the tap root surface  
270 injured). The proportion of plants with root injuries due to *D. radicum* larvae feeding increased  
271 linearly with the number of eggs deposited ( $\chi^2 = 13.0$ ;  $df = 1$ ;  $P < 0.001$ ; Fig. 3).

272 All plants produced a broccoli head, with an overall mean yield of  $295.9 \pm 13.8$ g per  
273 plant. Plant production was negatively and linearly correlated with the amount of eggs deposited  
274 on the plant ( $\chi^2 = 11.1$ ,  $df = 1$ ,  $P < 0.001$ ; Fig. 3). For a rise from 0 to 100 eggs, the predicted  
275 drop in broccoli mass was 136.1 g, for an attainable mass of 346.8 g, i.e. a loss of 39% of the  
276 attainable mass.

#### 277 **Discussion**

278 The two main results of this study are that 1) all plant development and growth traits, as  
279 well as the final mass of broccoli heads, were negatively correlated, at least transitorily, with  
280 *D. radicum* egg density and 2) no plants died and all produced a harvestable head despite a  
281 maximum pest density expected to lead to plant death.

282

#### 283 *D. radicum affects all development and growth traits and reduces plant production*

284

285 All development and growth traits were negatively and linearly correlated with pest  
286 density, at least for one measurement date. Additionally, broccoli mass decreased linearly with  
287 the amount of eggs deposited, when the first plants were ready for harvest. Our results therefore  
288 confirm that development delays and reduced plant growth during the growing season can lead  
289 to significant drops in head mass at harvest, at least when all plants are harvested in a row  
290 (Brandelero et al. 2016). This result is interesting because plant mortality is currently regarded  
291 as the main damage caused by the cabbage root fly on broccoli (El Titi 1979; Estorgues 2005).  
292 Similarly, El Titi (1977) found a negative correlation between *D. radicum* pupae density and  
293 vegetable mass in cauliflower, suggesting that this type of relationship between pest density  
294 and yield might be found for other *Brassica* crops. Our results indicate that (at least when no  
295 plant mortality occurs) the impact of *D. radicum* on plants is linear, with no threshold: even the  
296 lowest pest densities limit plant growth and head mass. This is not exactly consistent with the  
297 findings of Bligaard (1999), which suggested threshold effects in the relationship between the  
298 biomass (a growth trait) of cauliflowers and *D. radicum* egg density. Our results therefore  
299 suggest that *D. radicum* can be harmful even when it does not kill the plants.

300 However, all development and growth traits, except above ground plant height, were no  
301 longer correlated with pest density at the time of the final measurement. Thus it appears that  
302 the plants were compensating for the injuries caused by the pest. We therefore suggest that the  
303 production potential of each plant, i.e. the attainable yield if every plant had been harvested at  
304 individual maturity, may not have been modified by pest density in our experimental conditions,  
305 but that the plants had not yet fully compensated for the pest attack by the end of our experiment.  
306 The plants may thus have been able to tolerate the pest attacks (Strauss and Agrawal 1999;



307 Verdugo et al. 2016), as shown in many non-crop plants (e.g. Heichel and Turner 1983; Fowler  
308 and Rausher 1985; Karban and Courtney 1987; Maschinski and Whitham 1989) but also for  
309 instance in *Aphis gossypii* infesting cotton (Rosenheim et al. 1997). In the latter study,  
310 comparing infested and non-infested plants, the authors showed that leaf area was transitorily  
311 reduced by 58% when plants were infested with aphids but then by harvest the cotton plants  
312 had fully recovered so that yield was not affected (Rosenheim et al. 1997). In our crop/pest  
313 system, the consequences of pest infestation in terms of damage are likely to depend on the  
314 production situation. If the harvest is spread over several weeks, e.g. in the context of market  
315 gardening where produce is harvested only in time for sale, plants may reach their potential and  
316 *D. radicum* infestation may not affect final yield. In contrast, when the aim is to reduce the  
317 number of harvests to limit the costs, e.g. in the context of field vegetable production where the  
318 volumes are large and the associated costs for harvest are high, *D. radicum* is likely to add to  
319 the natural variability of the time needed to reach plant maturity (Dufault 1997; Grevsen 2000;  
320 Lindemann-Zutz et al. 2016). Thus, an infestation may lead to either additional harvests being  
321 performed, thereby increasing production costs, or to an increased amount of unharvested plants  
322 (because not fully mature at harvest), decreasing the income.

323

#### 324 *The absence of plant mortality and D. radicum mortality*

325

326 A surprising result of our experiment is that no plants died. Two hypotheses may explain  
327 this: 1) the maximum egg densities were not high enough to kill plants in our growing  
328 conditions or 2) the maximum egg densities should have killed the plants but the unexpected  
329 high developmental mortality of *D. radicum* led to an underestimation of the real harmfulness  
330 of this pest. In previous studies on cauliflower, El Titi (1979) and Bligaard (1999) reported  
331 significant plant mortality with much lower egg densities (respectively 60 and 25 eggs per  
332 plant), introduced when plants were at a similar developmental stage (~ 4 leaves). However,  
333 the link between pest density and damage is also likely to depend on plant growing conditions.  
334 Generally, good growing conditions increase plant compensating abilities (Fenemore 1984;  
335 Pedigo et al. 1986; Maschinski and Whitham 1989). More specifically, as root-feeding pests  
336 induce symptoms similar to those of drought (Gange and Brown 1989; Foggo and Speight  
337 1993), the availability of water may be a crucial factor determining the ability of plants to  
338 tolerate a pest attack (Godfrey and Yeargan 1985; Dunn and Frommelt 1998). In the present  
339 study, the spring was particularly wet: the meteorological station nearby (1.6km) recorded  
340 181.5mm throughout the experiment. Such precipitation levels are very unlikely to induce water  
341 stress in broccoli: in warmer conditions, Erdem et al. (2010) showed significant water stress  
342 only when spring broccoli received as little as 130mm, but no stress when it received 193mm  
343 or more. In addition, the windbreak effect of the cages probably limited desiccation. These  
344 favorable conditions may partly explain the absence of mortality.

345 Although the level of *D. radicum* mortality is usually high (~ 80-90%; Hughes and  
346 Salter 1959; Meyling et al. 2013), it was extreme in our experiment as a hundred percent  
347 mortality has never been reported. First, it cannot be ruled out that some pupae were beyond  
348 the area covered by the auger used (i.e. 12cm in diameter). Following Hughes (1960) and Finch  
349 et al. (1978) the area prospected was enough to collect more than 70% of the pupae. Given that  
350 we collected only two pupae within this area, it is unlikely that the pupae missed would have

351 substantially changed the estimation of *D. radicum* mortality rate. Then, some mortality  
352 occurred during the egg stage: we showed that between 10 and 25% of the eggs were unviable,  
353 which is consistent with previous studies conducted with this biological model (Neveu et al.  
354 1997). Actual egg mortality in the field could have been higher due to one supplementary  
355 handling, to deposit the eggs at the base of plant stems. Additional egg mortality could be  
356 attributable to predation by ground dwelling arthropods. Indeed, Fig. S1 in Supplementary  
357 materials shows that our exclusion setup did not completely suppress the ground dwelling  
358 arthropod fauna, at least during the weeks of plant infestation. The carabid family, that  
359 dominated this fauna, was essentially represented by two species *Metallina lampros* and *Phyla*  
360 *obtusa*. These share functional traits which likely determine the efficiency of ground dwelling  
361 predators on *D. radicum*: small body size and carnivorous diet (Purtauf et al. 2005). *Metallina*  
362 *lampros* is thought to be specialized on *D. radicum* and to contribute largely to its natural  
363 regulation (Hughes 1959; Coaker and Williams 1963; Andersen et al. 1983).

364 On the other hand, the results discussed above concerning the decreased plant growth  
365 and yield show that at least some of the eggs survived and produced harmful larvae. As almost  
366 no pupae were found around broccoli roots, some pest mortality also appears to have occurred  
367 during the larval stage. Based on previous studies performed in open fields, predation by ground  
368 dwelling arthropods on buried materials (here, *D. radicum* larvae) seems rather unlikely (Finch  
369 and Elliott 1994; Lee and Edwards 2012). However, using insect-proof cages meant that any  
370 trapped ground dwelling predators had virtually no alternative resources on which to prey  
371 besides the *D. radicum* eggs (no other *Brassica* herbivores were observed inside the cages). We  
372 also recorded high numbers of ants at the beginning of May (Fig. S1, in Supplementary  
373 materials), i.e. during larval development of the first introduced eggs. Several authors showed  
374 that many ant species are significant predators of below ground pests (Carroll and Janzen 1973;  
375 Yadav et al. 2012; Pacheco et al. 2017). For instance, Yadav et al. (2012) showed that ants  
376 caused 60% mortality of *Galleria mellonella* larvae that were being used as below ground  
377 sentinel prey. The unusually high mortality recorded here may therefore be related to the  
378 unusual presence of ants. Also, as mentioned above, the spring when the study took place was  
379 particularly wet and the soil was hydromorphic. High soil moisture is known to decrease larval  
380 survival (Finch and Skinner 1988) and could have contributed to the high mortality observed.  
381 Finally, the favorable growing conditions in our experiment may have increased the constitutive  
382 and induced resistance mechanisms of the plant and caused additional larval mortality.  
383 Induction of plant defense following infestation may explain some of the *D. radicum* mortality  
384 as well as some of the effects on plant traits, through a trade-off between resistance (i.e.  
385 mobilizing energy and matter to produce defensive compounds) and growth, i.e. an allocation  
386 cost (Strauss et al. 2002).

### 387 388 *Conclusion*

389  
390 In our experiment, the introduced *D. radicum* suffered high mortality rates, thus we could not  
391 establish a clear relationship between pest density and plant mortality. As a systematic  
392 insecticide treatment at planting (Spinosad, authorized in conventional and organic production)  
393 is currently used to prevent plant mortality, the re-assessment of this relationship is urgently  
394 needed. However, we showed that plant mortality is not the only cause of damage in this

395 crop/pest system and that sublethal effects can lead to a 40% decrease in broccoli mass at  
396 harvest even in conditions where the survival of the pest is not optimal. Based on our findings  
397 concerning pest harmfulness coupled to forecast models of population dynamics (e.g. Collier  
398 et al. 1991) it should be possible to develop decision support tools evaluating the relevance of  
399 the treatment, based on the expected pest infestation and its expected impact on crops. As  
400 predation by ground dwelling arthropods was probably a mortality factor of prime significance  
401 in the present study, it may be very valuable to take them into account in such models.

#### 402 **Acknowledgements**

403 We thank Paul Adam for helpful contribution in data collection, the “*Unité Expérimentale de*  
404 *la Motte*” (UE 0787) for the setup of the experimental plot and Dr. Leigh Gebbie for the English  
405 revision of this manuscript.

#### 406 **Author contribution statement**

407 All authors conceived and designed research. MV, LD and XM performed the field work. XM  
408 analyzed data. XM, ALR, AMC, VF and YT were involved in writing of the manuscript.

#### 409 **Funding**

410 This work was supported by the *GIS PIClé*g (grant n° P00315), the *Région Bretagne* (grant n°  
411 0461/COH14000/00001042) and the *Chaire AEI* (grant n° 2016 0190).

#### 412 **Conflict of interest**

413 The authors declare that they have no conflict of interest.

#### 414 **Compliance with ethical standards**

415 All applicable international, national, and/or institutional guidelines for the care and use of  
416 animals were followed.

417

418 **References**

- 419 Andersen A, Hansen ÅG, Rydland N, Øyre G (1983) Carabidae and Staphylinidae (Col.) as  
420 predators of eggs of the turnip root fly *Delia floralis* Fallén (Diptera, Anthomyiidae) in  
421 cage experiments. J Appl Entomol 95:499–506. doi: 10.1111/j.1439-  
422 0418.1983.tb02673.x
- 423 Aubertot J-N, Robin M-H (2013) Injury Profile Simulator, a qualitative aggregative modelling  
424 framework to predict crop injury profile as a function of cropping practices, and the  
425 abiotic and biotic environment. I. Conceptual bases. PLOS ONE 8:e73202. doi:  
426 10.1371/journal.pone.0073202
- 427 Bates D, Mächler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using  
428 lme4. J Stat Softw 67:1–48. doi: 10.18637/jss.v067.i01
- 429 Bligaard J (1999) Damage thresholds for cabbage root fly [*Delia radicum* (L.)] in cauliflower  
430 assessed from pot experiments. Acta Agric Scand Sect B — Soil Plant Sci 49:57–64.  
431 doi: 10.1080/09064719950135713
- 432 Bligaard J, Meadow R, Nielsen O, Percy-Smith A (1999) Evaluation of felt traps to estimate  
433 egg numbers of cabbage root fly, *Delia radicum*, and turnip root fly, *Delia floralis* in  
434 commercial crops. Entomol Exp Appl 90:141–148. doi: 10.1046/j.1570-  
435 7458.1999.00432.x
- 436 Boica Junior AL, Costa EN, Sardinha de Souza BH, et al (2015) Antixenosis and tolerance to  
437 *Diabrotica speciosa* (Coleoptera: Chrysomelidae) in common bean cultivars. Fla  
438 Entomol 98:464–472. doi: 10.1653/024.098.0212
- 439 Brandelero FD, Brum B, Storck L, et al (2016) Plant characters of broccoli determinants of  
440 head production. Cienc Rural 46:963–969. doi: 10.1590/0103-8478cr20150750
- 441 Cardona C, Gonzalez R, Schoonhoven AV (1982) Evaluation of damage to common beans by  
442 larvae and adults of *Diabrotica balteata* and *Cerotoma facialis*. J Econ Entomol  
443 75:324–327. doi: 10.1093/jee/75.2.324
- 444 Cargnelutti Filho A, Toebe M, Alves BM, et al (2015) Leaf area estimation of canola by leaf  
445 dimensions. Bragantia 74:139–148. doi: 10.1590/1678-4499.0388
- 446 Carroll CR, Janzen DH (1973) Ecology of foraging by ants. Annu Rev Ecol Syst 4:231–257.  
447 doi: 10.1146/annurev.es.04.110173.001311
- 448 Coaker TH, Williams DA (1963) The importance of some Carabidae and Staphylinidae as  
449 predators of the cabbage root fly, *Erioischia Brassicae* (Bouché). Entomol Exp Appl  
450 6:156–164. doi: 10.1111/j.1570-7458.1963.tb00613.x
- 451 Collier RH, Finch S, Phelps K (1991) A simulation model for forecasting the timing of attacks  
452 of *Delia radicum* on cruciferous crops. EPPO Bull 21:419–424. doi: 10.1111/j.1365-  
453 2338.1991.tb01271.x
- 454 Desneux N, Decourtye A, Delpuech J-M (2007) The sublethal effects of pesticides on beneficial  
455 arthropods. Annu Rev Entomol 52:81–106. doi:  
456 10.1146/annurev.ento.52.110405.091440
- 457 Devictor V, Clavel J, Julliard R, et al (2010) Defining and measuring ecological specialization.  
458 J Appl Ecol 47:15–25. doi: 10.1111/j.1365-2664.2009.01744.x
- 459 Dossdall LM, Herbut MJ, Cowle NT (1994) Susceptibilities of species and cultivars and canola  
460 and mustard to infestation by root maggots (*Delia* spp.) (Diptera, Anthomyiidae). Can  
461 Entomol 126:251–260

- 462 Dufault RJ (1997) Determining heat unit requirements for broccoli harvest in coastal south  
463 carolina. *J Am Soc Hortic Sci* 122:169–174
- 464 Dunn JP, Frommelt K (1998) Effects of below-ground herbivory by *Diabrotica virgifera*  
465 *virgifera* (Col., Chrysomelidea) and soil moisture on leaf gas exchange of maize. *J Appl*  
466 *Entomol* 122:179–183. doi: 10.1111/j.1439-0418.1998.tb01481.x
- 467 Eilenberg J, Hajek A, Lomer C (2001) Suggestions for unifying the terminology in biological  
468 control. *BioControl* 46:387–400. doi: 10.1023/A:1014193329979
- 469 El Titi A (1977) Determination of the economic threshold of the cabbage root fly (*Erioischia*  
470 *brassicae* Bouché) in early cauliflower. I. The relationship between population density  
471 of the fly, chemical control and yield. *Z Pflanzenkrankh Pflanzenschutz* 84:65–77
- 472 El Titi A (1979) Further experiments on the determination of the economic threshold of the  
473 cabbage root fly (*Erioischia brassicae* Bouché) in early cauliflower. *Z Pflanzenkrankh*  
474 *Pflanzenschutz* 86:65–74  
475
- 476 Erdem Y, Arin L, Erdem T, et al (2010) Crop water stress index for assessing irrigation  
477 scheduling of drip irrigated broccoli (*Brassica oleracea* L. var. *italica*). *Agric Water*  
478 *Manag* 98:148–156. doi: 10.1016/j.agwat.2010.08.013
- 479 Estorgues V (2005) Maladies et ravageurs des légumes de plein champ en Bretagne. *Chambres*  
480 *d’agriculture de Bretagne*
- 481 Faraway JJ (2006) Extending the linear model with R : generalized linear, mixed effects and  
482 non parametric regression models. Chapman & Hall/CRC Taylor & Francis group
- 483 Feller C, Bleiholder H, Buhr L, et al (1995) Phänologische Entwicklungsstadien von  
484 Gemüsepflanzen. II. Fruchtgemüse und Hülsenfrüchte. *Nachrichtenblatt Dtsch*  
485 *Pflanzenschutzdienste* 47:193–206
- 486 Fenemore PG (1984) *Plant Pests and Their Control*. Elsevier, United Kingdom
- 487 Finch S, Elliott MS (1994) Predation of cabbage root fly eggs and larvae by carabid ground  
488 beetles - fact or fantasy? *IOBC-WPRS Bull* 17:109–114
- 489 Finch S, Skinner G (1988) Mortality of the immature stages of the cabbage root fly. In:  
490 *Proceedings of the CEC / IOBC experts’group meeting*. Cavalloro R. & Pellerents C.,  
491 Rennes, pp 45–48
- 492 Finch S, Skinner G, Freeman GH (1978) Distribution and analysis of cabbage root fly pupal  
493 populations. *Ann Appl Biol* 88:351–356
- 494 Foggo A, Speight MR (1993) Root damage and water stress: treatments affecting the  
495 exploitation of the buds of common ash *Fraxinus excelsior* L., by larvae of the ash bud  
496 moth *Prays fraxinella* Bjerk. (Lep., Yponomeutidae). *Oecologia* 96:134–138. doi:  
497 10.1007/BF00318041
- 498 Fowler NL, Rausher MD (1985) Joint effects of competitors and herbivores on growth and  
499 reproduction in *Aristolochia reticulata*. *Ecology* 66:1580–1587. doi: 10.2307/1938020
- 500 Fox J, Weisberg S (2011) *An R companion to applied regression*, 2nd edn. Sage, Thousand  
501 Oaks (CA)
- 502 Gange AC, Brown VK (1989) Effects of root herbivory by an insect on a foliar-feeding species,  
503 mediated through changes in the host plant. *Oecologia* 81:38–42. doi:  
504 10.1007/BF00377007

505 Geiger F, Bengtsson J, Berendse F, et al (2010) Persistent negative effects of pesticides on  
506 biodiversity and biological control potential on European farmland. *Basic Appl Ecol*  
507 11:97–105. doi: 10.1016/j.baae.2009.12.001

508 Godfrey LD, Yeagan KV (1985) Influence of soil moisture and weed density on clover root  
509 curculio, *Sitona hispidulus*, larval stress to alfalfa. *J Agric Entomol* 2:370–377

510 Grevsen K (2000) Modelling plant development of broccoli. *Acta Hort* 533:567–574. doi:  
511 10.17660/ActaHortic.2000.533.71

512 Heichel GH, Turner NC (1983) CO<sub>2</sub> assimilation of primary and regrowth foliage of red maple  
513 (*Acer rubrum* L.) and red oak (*Quercus rubra* L.): response to defoliation. *Oecologia*  
514 57:14–19. doi: 10.1007/BF00379555

515 Hopkins RJ, Birch ANE, Griffiths DW, et al (1995) Changes in the dry matter, sugar, plant  
516 fibre and lignin contents of swede, rape and kale roots in response to turnip root fly  
517 (*Delia floralis*) larval damage. *J Sci Food Agric* 69:321–328. doi:  
518 10.1002/jsfa.2740690308

519 Hughes RD (1960) A method of estimating the numbers of cabbage root fly pupae in the soil.  
520 *Plant Pathol* 9:15–17

521 Hughes RD (1959) The natural mortality of *Erioischia brassicae* (Bouché) (Diptera,  
522 Anthomyiidae) during the egg stage of the first generation. *J Anim Ecol* 28:343–357

523 Hughes RD, Salter DD (1959) Natural mortality of *Erioischia brassicae* (Bouché) (Diptera,  
524 Anthomyiidae) during the immature stages of the first generation. *J Anim Ecol* 28:231–  
525 241

526 Hunter MD (2001) Out of sight, out of mind: the impacts of root-feeding insects in natural and  
527 managed systems. *Agric For Entomol* 3:3–9. doi: 10.1046/j.1461-9563.2001.00083.x

528 International conference on global crop losses (2017) Synthesis and report. INRA, Paris

529 IUSS working group WRB (2015) World reference base for soil resources 2014, update 2015 -  
530 International soil classification system for naming soils and creating legends for soil  
531 maps. FAO, Rome

532 Josso C, Le Ralec A, Raymond L, et al (2013) Effects of field and landscape variables on crop  
533 colonization and biological control of the cabbage root fly *Delia radicum*. *Landsc Ecol*  
534 28:1697–1715. doi: 10.1007/s10980-013-9928-3

535 Karban R, Courtney S (1987) Intraspecific host plant choice: lack of consequences for  
536 *Streptanthus tortuosus* (Cruciferae) and *Euchloe hyantis* (Lepidoptera: Pieridae). *Oikos*  
537 48:243–248. doi: 10.2307/3565508

538 Kloen H, Altieri MA (1990) Effect of mustard (*Brassica hirta*) as a non-crop plant on  
539 competition and insect pests in broccoli (*Brassica oleracea*). *Crop Prot* 9:90–96. doi:  
540 10.1016/0261-2194(90)90084-K

541 Lamy F, Dugravot S, Cortesero AM, et al (2017) One more step toward a push-pull strategy  
542 combining both a trap crop and plant volatile organic compounds against the cabbage  
543 root fly *Delia radicum*. *Environ Sci Pollut Res* 1–12. doi: 10.1007/s11356-017-9483-6

544 Lee JC, Edwards DL (2012) Impact of predatory carabids on below- and above-ground pests  
545 and yield in strawberry. *BioControl* 57:515–522. doi: 10.1007/s10526-011-9425-z

546 Lenth RV (2017) emmeans: Estimated Marginal Means, aka Least-Squares Means

547 Lindemann-Zutz K, Fricke A, Stützel H (2016) Prediction of time to harvest and its variability  
548 in broccoli (*Brassica oleracea* var. *italica*) Part I. Plant developmental variation and

549 forecast of time to head induction. *Sci Hortic* 198:424–433. doi:  
550 10.1016/j.scienta.2015.12.023

551 Maschinski J, Whitham TG (1989) The continuum of plant responses to herbivory: the  
552 influence of plant association, nutrient availability, and timing. *Am Nat* 134:1–19. doi:  
553 10.1086/284962

554 Meyling NV, Navntoft S, Philipsen H, et al (2013) Natural regulation of *Delia radicum* in  
555 organic cabbage production. *Agric Ecosyst Environ* 164:183–189. doi:  
556 10.1016/j.agee.2012.09.019

557 Murray PJ, Hatch DJ, Cliquet JB (1996) Impact of insect root herbivory on the growth and  
558 nitrogen and carbon contents of white clover (*Trifolium repens*) seedlings. *Can J Bot*  
559 74:1591–1595. doi: 10.1139/b96-192

560 Neveu N, Langlet X, Brunel E, et al (1997) The fine structure of the egg shells of the cabbage  
561 maggot, *Delia radicum* L. (Diptera: Anthomyiidae) and its relation with developmental  
562 conditions and oviposition site. *Can J Zool* 75:535–541

563 Oerke E-C (2006) Crop losses to pests. *J Agric Sci* 144:31–43. doi:  
564 10.1017/S0021859605005708

565 Olfati JA, Peyvast G, Shabani H, Nosratie-Rad Z (2010) An estimation of individual leaf area  
566 in cabbage and broccoli using non-destructive methods. *J Agric Sci Technol* 12:627–  
567 632

568 Pacheco R, Camacho GP, Frizzo TLM, Vasconcelos HL (2017) Effects of land-use changes on  
569 ecosystem services: decrease in ant predation in human-dominated landscapes in central  
570 Brazil. *Entomol Exp Appl* 162:302–308. doi: 10.1111/eea.12542

571 Pedigo LP, Hutchins SH, Higley LG (1986) Economic injury levels in theory and practice.  
572 *Annu Rev Entomol* 31:341–368. doi: 10.1146/annurev.en.31.010186.002013

573 Poston FL, Pedigo LP, Welch SM (1983) Economic injury levels: reality and practicality. *Bull*  
574 *Entomol Soc Am* 29:49–53. doi: 10.1093/besa/29.1.49

575 Purtauf T, Dauber J, Wolters V (2005) The response of carabids to landscape simplification  
576 differs between trophic groups. *Oecologia* 142:458–464. doi: 10.1007/s00442-004-  
577 1740-y

578 R core team (2017) R: a language and environment for statistical computing. R foundation for  
579 statistical computing, Vienna, Austria

580 Rogers DJ, Brier HB (2010) Pest-damage relationships for *Helicoverpa armigera* (Hübner)  
581 (Lepidoptera: Noctuidae) on vegetative soybean. *Crop Prot* 29:39–46. doi:  
582 10.1016/j.cropro.2009.08.016

583 Rosenheim JA, Wilhoit LR, Goodell PB, et al (1997) Plant compensation, natural biological  
584 control, and herbivory by *Aphis gossypii* on pre-reproductive cotton: the anatomy of a  
585 non-pest. *Entomol Exp Appl* 85:45–63. doi: 10.1046/j.1570-7458.1997.00233.x

586 Strauss SY, Agrawal AA (1999) The ecology and evolution of plant tolerance to herbivory.  
587 *Trends Ecol Evol* 14:179–185. doi: 10.1016/S0169-5347(98)01576-6

588 Strauss SY, Rudgers JA, Lau JA, Irwin RE (2002) Direct and ecological costs of resistance to  
589 herbivory. *Trends Ecol Evol* 17:278–285. doi: 10.1016/S0169-5347(02)02483-7

590 Tartaglia F de L, Righi EZ, da Rocha L, et al (2016) Non-destructive models for leaf area  
591 determination in canola. *Rev Bras Eng Agric E Ambient* 20:551–556. doi:  
592 10.1590/1807-1929/agriambi.v20n6p551-556

- 593 Teixeira MLF, Coutinho LCH, Franco AA (1996) Effects of *Cerotoma arcuata* (Coleoptera:  
594 Chrysomelidae) on predation of nodules and on N<sub>2</sub> fixation of *Phaseolus vulgaris*. J  
595 Econ Entomol 89:165–169. doi: 10.1093/jee/89.1.165
- 596 Turnock W, Timlick B, Galka B, Palaniswamy P (1992) Root maggot damage to canola and  
597 the distribution of *Delia* spp. (Diptera, Anthomyiidae) in Manitoba. Can Entomol  
598 124:49–58
- 599 Verdugo JA, Francis F, Ramirez CC (2016) A review on the complexity of insect-plant  
600 interactions under varying levels of resources and host resistance: the case of *Myzus*  
601 *persicae*-*Prunus persica*. Biotechnol Agron Soc Environ 20:533–541
- 602 Yadav P, Duckworth K, Grewal PS (2012) Habitat structure influences below ground biocontrol  
603 services: A comparison between urban gardens and vacant lots. Landsc Urban Plan  
604 104:238–244. doi: 10.1016/j.landurbplan.2011.10.018
- 605 Zadoks JC (1985) On the conceptual basis of crop loss assessment: the threshold theory. Annu  
606 Rev Phytopathol 23:455–473. doi: 10.1146/annurev.py.23.090185.002323
- 607



608 **Table 1** Pattern of artificial infestation of the broccoli plants with cabbage root fly eggs  
 609

1 <sup>st</sup> infestation (3 weeks after planting)	0	1	3	6	9	12	15	18	24	30
2 <sup>nd</sup> infestation (4 weeks after planting)	0	3	4	8	12	16	20	24	32	40
3 <sup>rd</sup> infestation (5 weeks after planting)	0	1	3	6	9	12	15	18	24	30
Total	0	5	10	20	30	40	50	60	80	100

610

611 **Table 2** Effect of the number of *D. radicum* eggs deposited per plant and its interaction with  
 612 the date of measurement on the development and growth traits recorded for the broccoli plants.  
 613 Bold p-values show significant differences at  $\alpha = 5\%$   
 614

	Number of eggs				Interaction “number of eggs: date”		
	$\chi^2$	DF	P-val	Trend	$\chi^2$	DF	P-val
Number of leaves	6.61	1	<b>0.010</b>	↘	15.17	9	0.086
Proportion of plants with at least one lateral shoot	2.36	1	0.125		8.64	5	0.124
Proportion of plants with a visible inflorescence	8.25	1	<b>0.004</b>	↘	2.34	1	0.126
Relative largest leaf area	13.66	1	<b>&lt; 0.001</b>	↘	24.16	8	<b>0.002</b>
Height above ground of the apical meristem	5.09	1	<b>0.024</b>	↘	54.82	8	<b>&lt; 0.001</b>

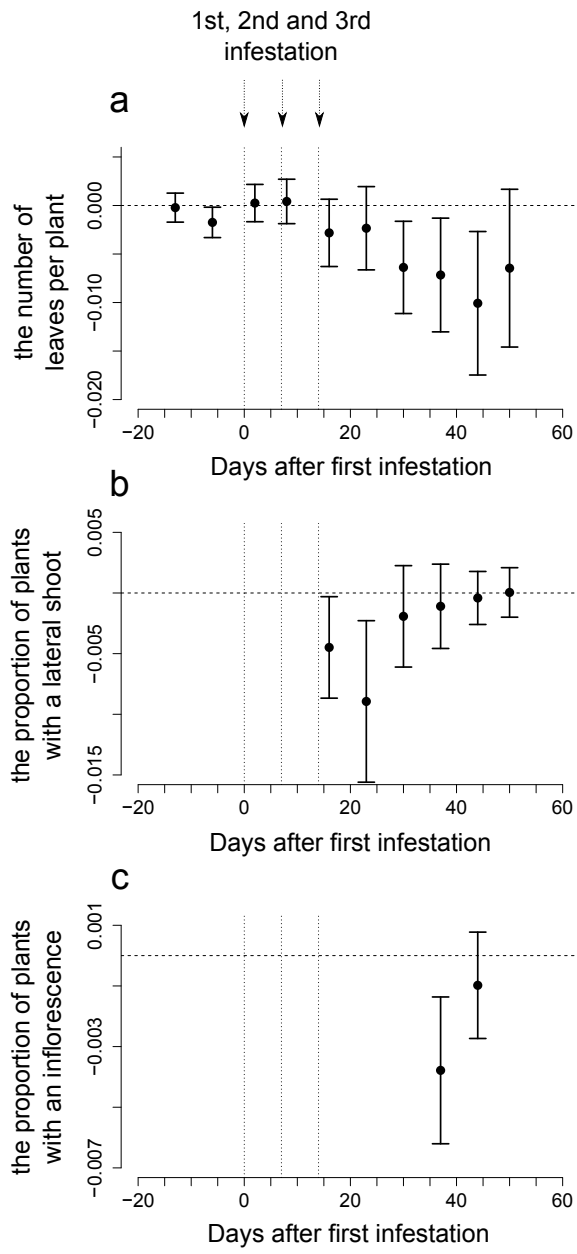
615

616 **Fig. 1** Evolution through time of the linear trend between plant traits (a: number of leaves; b:  
617 proportion of plants with at least one lateral shoot; c: proportion of plants with an inflorescence;  
618 d: height above ground of the apical meristem in cm and e: relative largest leaf area in cm<sup>2</sup>) and  
619 the number of eggs deposited. Each point and associated error bar represents the estimate  $\pm$   
620 95% confidence interval of this trend, obtained with function ‘emtrends’ (Lenth 2017). A  
621 negative value indicates that plant development or growth is negatively correlated with the  
622 number of eggs deposited at the time of measurement. The trend is significant (i.e. the effect of  
623 the number of eggs on plant trait is significant at a given time) if the error bar does not cross  
624 the horizontal dashed line in 0. Vertical dotted lines represent the three artificial infestations of  
625 broccoli plants

626  
627 **Fig. 2** Plant development (a: leaf number, mean  $\pm$  SE; b: proportion of plants with at least one  
628 lateral shoot, prop  $\pm$  SE and c: proportion of plants with a visible inflorescence, prop  $\pm$  SE) and  
629 growth traits (d: height above ground of the apical meristem in cm, mean  $\pm$  SE; e: relative  
630 largest leaf area in cm<sup>2</sup>, mean  $\pm$  SE) as a function of the number of *D. radicum* eggs deposited  
631 per plant for the measurement date indicated above the graphs, corresponding to the time when  
632 the correlation between plant trait and egg density was the most negative (Fig. 1). The dashed  
633 grey lines present the regression curves obtained with the coefficients of the models presented  
634 in the text and in Table 2

635  
636 **Fig. 3** Proportion of plants showing root injuries (prop  $\pm$  SE; a) and broccoli mass at harvest (in  
637 grams, mean  $\pm$  SE; b) according to the number of *D. radicum* eggs deposited per plant

Estimated slope of the relationship between the number of eggs deposited and



Estimated slope of the relationship between the number of eggs deposited and

