

1 Original article

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3 **Morphological adaptation of a palatable plant to long-term grazing can shift interactions**  
4 **with an unpalatable plant from facilitative to competitive.**

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19 Running headline: Changes in plant interactions in a grazed habitat

20

21 **Abstract**

22 Unpalatable plants can protect palatable neighbor plants from grazing pressure, but  
23 morphological evolution of a palatable species might change its interactions with unpalatable  
24 plants. We predicted that when a palatable species has locally adapted to grazing by expressing  
25 a dwarf phenotype that reduces grazer accessibility, the dwarf plants experience relatively more  
26 competitive effects than facilitative effects from large, well-defended, unpalatable species. We  
27 used a transplant experiment, in which both dwarf and large ecotypes of a palatable annual  
28 species, *Persicaria longiseta*, were transplanted outside and inside the canopy of an unpalatable  
29 nettle, *Urtica thunbergiana*, in a long-term deer grazing habitat of Nara Park, Japan. The dwarf  
30 ecotype of *Persicaria* has adapted to the grazing environments of the park by exhibiting  
31 inherently short shoots and small leaves, whereas the large ecotype is found in habitats with no  
32 grazing history. A previous common-garden study suggested that the phenotypic differences  
33 were genetically based and that phenotypic plasticity contributed little to the morphological  
34 difference. The large-phenotype of *Persicaria* experienced significantly increased  
35 morphological size, survival, and reproductive output under the *Urtica* canopy compared to  
36 outside the canopy, whereas these traits of the dwarf phenotype were reduced under the *Urtica*  
37 canopy compared to outside. These results indicate that the net effects of *Urtica* on *Persicaria*  
38 were positive for the large ecotype and negative for the dwarf ecotype. Thus, the morphological  
39 adaptation of a palatable species to avoid grazing altered its interactions with a large,  
40 well-defended neighbor.

41

42 **Key words:** competition, dwarf phenotype, facilitation, large phenotype, long grazing history,  
43 *Persicaria longiseta*

44

45 **INTRODUCTION**

46 Facilitative interactions between plant species play an important role in determining the spatial  
47 distributions of species, community structures, and ecosystem functioning (Callaway 1997;  
48 Eccles et al. 1999; Callaway et al. 2000; Tirado and Pugnaire 2003; Callaway et al. 2005;  
49 Manier and Hobbs 2006; Osem et al. 2007), and are effective in conserving the biodiversity of  
50 plant communities (Callaway et al. 2000; Bossuyt et al. 2005; Callaway et al. 2005).  
51 Facilitation can also act as an evolutionary force, such as in the convergence of species' niches  
52 (reviewed in Brooker et al. 2008). However, few studies have examined the evolutionary  
53 aspects of facilitation (Bronstein 2009; Michalet et al. 2011). For example, whether facilitative  
54 interactions are maintained or break down over a long-term evolutionary period has been little  
55 explored.

56         The stress gradient hypothesis predicts that increased environmental severity increases  
57 facilitative (positive) interactions (Brooker et al. 2006; Graff et al. 2007; Smit et al. 2007;  
58 Alberti et al. 2008). On the other hand, stressful environments likely favor genotypes and  
59 species with stress-tolerant life history traits. Therefore, when plants evolve stress-tolerant  
60 traits in stressful habitats, plant–plant interactions in these habitats appear to mainly occur  
61 between stress-tolerant plants. Liancourt et al. (2005) and Maestre et al. (2009) suggested that  
62 the effect of neighbors can become negative when both interacting species have similar  
63 “stress-tolerant” life histories.

64         It is well recognized that unpalatable species exert facilitative effects on their palatable  
65 neighbors by protecting them from grazers in grazed communities (Oesterheld and Oyarzabal  
66 2004; Russell and Fowler 2004; Bossuyt et al. 2005). Meanwhile, a long evolutionary history of  
67 grazing often selects for small plant phenotypes (Detling and Painter 1983; McKinney and

68 Fowler 1991; McNaughton 1984; Polley and Detling 1988; Fahnestock and Detling 2000;  
69 Kotanen and Bergelson 2000; Ishikawa et al. 2006; Suzuki 2008; Suzuki et al. 2009). While the  
70 small plants experience reduced grazing damage, they tend to have reduced competitive  
71 abilities (Briske and Anderson 1992; Hartvigsen and McNaughton 1995; Osem *et al.* 2004;  
72 McGuire and Agrawal 2005). Therefore, when both an unpalatable species and a palatable  
73 species are adapted to grazing in different ways, competitive rather than positive, facilitative  
74 interactions between them may be increased (Baraza et al. 2006; Callaway et al. 2005; Suzuki  
75 and Suzuki 2011a; Vandenberghe et al. 2009). Our previous study demonstrated that a palatable  
76 species adapted to grazing by expressing a dwarf phenotype experienced little effects of  
77 facilitation by a large unpalatable species (Suzuki and Suzuki 2011a, b). Therefore, we predict  
78 that the net effects of unpalatable plants on palatable plants shift from positive to negative when  
79 the phenotypes of palatable plants evolve from large morphology to small morphology. Plants  
80 that have evolved small phenotypes can survive alone in grazed habitats, but experience  
81 competition rather than facilitation from large unpalatable species.

82 To test this hypothesis, we conducted a transplant experiment in which both dwarf and  
83 large phenotypes of a palatable annual species, *Persicaria longiseta*, were transplanted outside  
84 and inside the canopy of an unpalatable nettle, *Urtica thunbergiana*, in a long-term deer grazing  
85 habitat of Nara Park, Japan. The population of *P. longiseta* in Nara Park exhibits an inherently  
86 dwarf morphology with shorter shoots and smaller leaves as a consequence of a local  
87 adaptation to grazing (Suzuki 2008). Conversely, populations of *Persicaria* with no history of  
88 grazing exhibit inherently large growth forms (Suzuki 2008). We compared the performances  
89 (growth, survival, and reproduction) of transplanted individuals between dwarf and  
90 large-phenotypes of *Persicaria* outside and inside the *Urtica* canopy during a growing season.

91 We predicted that under the *Urtica* canopy, the dwarf phenotype of *Persicaria* would  
92 experience relatively more competitive effects from *Urtica* while the large phenotype would  
93 experience facilitative effects.

94

## 95 **MATERIALS AND METHODS**

### 96 ***Study species***

97 We examined the interactions between a palatable species, *Persicaria longiseta* (De Bruyn)  
98 Kitag., and an unpalatable species, *Urtica thunbergiana* Siebold et Zucc. *Persicaria longiseta*  
99 is an annual herb that is distributed widely throughout East Asia and is common to roadsides,  
100 farms, edges of rice fields, and gardens in Japan. Seed germination occurs primarily from the  
101 end of March to April. Flowering is initiated in July. Reproductive plants produce a large  
102 number of small seeds (ca. 1.5 mg per seed) but do not regenerate vegetatively. Fruit maturation  
103 and seed dispersal occur from July to December, and seeds are dispersed by gravity as soon as  
104 they mature. After reproduction, the plant dies by mid December.

105 The nettle, *Urtica thunbergiana* is a perennial herb distributed within forest understories  
106 throughout central and southern Japan. This species possesses stinging hairs containing toxins  
107 on its stems and both surfaces of leaves; thus, they are unpalatable to deer because of these  
108 physical and chemical defenses (Kato et al. 2008). During a growing season, plants develop  
109 numerous shoots proximal to the base. Most aboveground parts wither during the winter season  
110 (December–March).

111

### 112 ***Seed collection***

113 To cultivate plants for our transplant experiment, seeds of *Persicaria* were collected from

114 two sites in Nara City: Nara Park and Heijo Ruins (hereafter referred to as the Nara and Heijo  
115 sites, respectively). The current grazing regimes and grazing histories of the two sites were  
116 strikingly different; the Heijo site has no history of grazing, while the Nara site has both a long  
117 history of deer grazing and is currently grazed.

118 The Nara and Heijo sites are located at the eastern edge of Nara Prefecture, western Japan.  
119 The annual mean precipitation is 1333 mm, and the monthly mean temperatures range from  
120 3.8°C in January to 26.6°C in August in this region (for the years 1971–2000; Japan  
121 Meteorological Agency 2011). Sika deer (*Cervus nippon* Temminck) populations have been  
122 distributed in the park at the Nara site (34°41'N, 135°50'E) for more than 1,200 years, as they  
123 are protected for religious reasons. Nara park is 660 ha in area and includes open grasslands and  
124 evergreen forests. Individuals of *P. longiseta* are often scattered in grasslands and in the sparse  
125 understory of the tree stands in the park. Our common-garden experiment revealed that the  
126 population of *Persicaria* at the Nara site is an ecotype adapted to grazed environments and  
127 exhibits an inherently dwarf morphology with shorter shoots and smaller leaves compared to  
128 populations at the Heijo site, which has no grazing history (Suzuki 2008). Under cultivated  
129 conditions, the leaf and shoot lengths of the Nara population were 0.6 and 0.4 times smaller,  
130 respectively, than those of the Heijo population (Suzuki 2008). The population of *U.*  
131 *thunbergiana* in Nara Park has also evolved a much higher density of stinging hairs compared  
132 to those found in ungrazed areas (Kato et al. 2008).

133 The Heijo site (34°41'N, 135°47'E) is 120 ha in area and located 4 km west of the Nara site.  
134 No current or historical grazing by large herbivores has been observed at the site. *P. longiseta*  
135 individuals at the Heijo site are often distributed in irrigation ditches.

136 To cultivate *Persicaria* plants from seeds, in November and December 2007, seeds were

137 collected from 25 randomly selected reproductive *Persicaria* plants at each site within an area  
138 of 100-m radius. Reproductive plants were selected at least 1 m apart to insure that they were  
139 different individuals. Collected seeds of each reproductive plant were separately stored at room  
140 temperature before the transplant experiment.

141

## 142 **Transplant experiment**

143 Our transplant experiment was carried out with 100 *Persicaria* plants in Nara Park.  
144 Prior to transplanting, plants were cultivated from seeds at a common garden at the Nara  
145 University of Education in Nara City (34°40'N, 135°50'E). Four seeds per mother plant were  
146 incubated at 4°C under humid conditions for 12 days from 7 April 2008. After the moist chilling  
147 treatment, seeds were sown in Jiffy pots (5 × 5 cm, 5 cm deep) filled with vermiculite, which  
148 were placed within the common garden. Seedlings were fertilized with 2,000 times dilution of  
149 Hyponex solution (6-10-5 NPK, HYPONeX JAPAN Corp., Ltd, Osaka, Japan) once a week.  
150 We selected two seedlings of similar size per mother plant for the transplant experiment.

151 The study area was established in a shady-moist environment in the understory of  
152 sparsely planted Japanese cedars, *Cryptomeria japonica*, within a long-term deer-grazed  
153 habitat in Nara Park. The densities of *Urtica* and *Persicaria* were high in the study area. Our  
154 previous studies revealed that *Persicaria* individuals naturally establish outside and inside the  
155 *Urtica* canopy in the study area, but *Persicaria* experience little benefit from facilitation by  
156 *Urtica* (Suzuki and Suzuki 2011a, b). We randomly selected 25 *Urtica* individuals within the  
157 study area. These individuals averaged 20–34 cm in height and 32–61 cm in diameter during  
158 the study period. On 26 May 2008, 5 weeks after the seeds were sown, *Persicaria* individuals  
159 from the Nara and Heijo populations were transplanted with Jiffy pots to positions inside and

160 outside the canopy of each *Urtica* (two populations  $\times$  two positions  $\times$  25 replicates; total  $N =$   
161 100). We assumed that the Jiffy pots had negligible effects on root growth of transplanted  
162 individuals and belowground competition between *Persicaria* and *Urtica* individuals because  
163 the Jiffy pots biodegraded shortly after the transplantation and plant roots had already begun to  
164 penetrate the pots before transplanting in the field. Hereafter, we refer to a set of four *Persicaria*  
165 individuals around a *Urtica* plant as a “block” so that it can be included as a random effect in the  
166 statistical analyses. Plants transplanted outside of the *Urtica* canopy were placed 50 cm from  
167 the canopy edge. Although these experimental positions inside and outside of the *Urtica* canopy  
168 were artificial, similar spatial relationships between *Persicaria* and *Urtica* were regularly  
169 observed under natural conditions (Suzuki and Suzuki 2011a, b). After transplanting, survival,  
170 plant height, number of leaves, and number of shoots were monitored for all plants at 27–35 day  
171 intervals during the growing season. We measured the maximum length and the perpendicular  
172 axis ( $D_1$  and  $D_2$ , respectively) of the area covered by aboveground parts of each plant at each  
173 census. As a measure of plant size, we calculated the relative area covered by each plant ( $D_1 \times$   
174  $D_2$ ). The number of flowers produced by each transplanted plant was counted in October. We  
175 also estimated the lifetime fitness of each plant as the number of flowers produced by the plant  
176 multiplied by the survival rate of plants in the treatment to which the plant belonged.

177

## 178 **Analysis**

179 Data analysis was conducted using R (ver. 2.12.2.; R Development Core Team 2011).  
180 We examined the effects of population, distance from *Urtica*, and their interaction on survival,  
181 plant morphological traits, and reproductive outputs of *Persicaria* individuals. Plant survival  
182 during the experimental periods was analyzed using the Cox proportional hazards regression

183 model containing a random effect (block) by the `coxme` function from the `kinship` library in R.  
184 The number of flowers in October and lifetime fitness were analyzed using a generalized linear  
185 mixed-effects model (GLMM) containing a random effect (block) (the `lmer` function from the  
186 `lme4` library in R). The number of flowers was analyzed with a Poisson error distribution, and  
187 lifetime fitness was analyzed with a normal error distribution after log-transformation. We also  
188 analyzed repeated measures of plant morphological traits using GLMMs (the `lmer` function in  
189 R). Height and plant size were analyzed with a normal error distribution after  
190 log-transformation, and the number of leaves and number of shoots were analyzed with a  
191 Poisson error distribution. All models included population, distance (inside or outside the  
192 *Urtica* canopy), and their interaction as fixed effects and block as a random effect. The GLMMs  
193 of morphological traits also included the date of measurement and individual identities as  
194 random effects. All GLMMs were conducted using restricted maximum likelihood estimation.  
195 *P* values for all GLMMs were calculated using the `pvals.fnc` function from the language R  
196 library in R. Back-transformed means and standard errors of the analyses are presented  
197 throughout.

198

## 199 **RESULTS**

200 We found interactive effects of population and distance from *Urtica* individuals on the  
201 performance of *Persicaria* individuals. Throughout all experimental periods except October,  
202 the number of surviving individuals from the Nara population exhibiting the dwarf phenotype  
203 was lower inside the *Urtica* canopy than outside of it, whereas that of the Heijo population with  
204 the large phenotype was higher inside the *Urtica* canopy (Fig. 1). In October, survival of  
205 individuals from the Heijo population did not differ with *Urtica* distance. The interaction

206 between population and distance from *Urtica* was significant (Table 1).

207 Plant height of *Persicaria* individuals tended to be lower inside the *Urtica* canopy for  
208 the Nara population, whereas that of the Heijo population tended to be higher inside the *Urtica*  
209 canopy than outside of it for most experimental periods (Fig. 2a). However, the interaction  
210 effect between population and distance from *Urtica* was marginally insignificant, and only the  
211 population effect was significant (Table 2). *Persicaria* individuals of both populations located  
212 inside the *Urtica* canopy tended to have fewer leaves than those located outside the canopy in  
213 most periods (Fig. 2b). The population effect and the interaction effect between population and  
214 distance from *Urtica* were statistically significant (Table 2). The number of shoots showed  
215 similar trends to the number of leaves (Fig. 2c). *Persicaria* individuals located inside the *Urtica*  
216 canopy had fewer shoots than individuals located outside the canopy irrespective of population.  
217 The effects of population and distance on the number of shoots were significant (Table 2).  
218 *Persicaria* individuals from the Nara population had smaller plant sizes inside than outside the  
219 *Urtica* canopy throughout all experimental periods. Individuals from the Heijo population were  
220 smaller inside the *Urtica* canopy than outside of it in June and September, but plant sizes were  
221 larger inside of the canopy in July, August, and October (Fig. 2d). The population effect was  
222 only significant on plant size, whereas the distance effect and the interaction effect were  
223 insignificant on plant size (Table 2).

224 Individuals of the Nara population produced fewer flowers inside of the *Urtica* canopy  
225 than outside of it, whereas individuals of the Heijo population produced more flowers inside the  
226 *Urtica* canopy (Fig. 3a). The interaction effect between population and distance was significant  
227 (Table 3). Lifetime fitness also showed a similar trend to the number of flowers (Fig. 3b). The  
228 population effect, the distance effect, and the interaction effect of population and distance from

229 *Urtica* on lifetime fitness were significant (Table 3).

230

## 231 **DISCUSSION**

232 The main finding of this study is that unpalatable large *Urtica* plants had negative effects on the  
233 grazing-adapted, dwarf-phenotype of *Persicaria*, whereas *Urtica* had positive effects on the  
234 grazing-sensitive, large-phenotype of *Persicaria*. *Urtica* positively affected the survival, height,  
235 plant size, and reproductive output of individuals from the Heijo population, but had a negative  
236 effect on individuals from the Nara population throughout the growing season. The Nara  
237 populations of *Persicaria* have locally adapted to grazing by expressing a dwarf phenotype that  
238 reduces grazer accessibility. However, the dwarf phenotype is likely competitively inferior to  
239 its larger counterparts (Briske and Anderson 1992; Hartvigsen and McNaughton 1995; Osem et  
240 al. 2004; McGuire and Agrawal 2005). The opposite effects of *Urtica* on the two ecotypes of  
241 *Persicaria* suggest that morphological evolution from the large to the dwarf-phenotype of  
242 *Persicaria* might have altered the interactions between *Persicaria* and *Urtica* from facilitation  
243 to competition in a grazed habitat.

244 Facilitative effects are often demonstrated in habitats with recent grazing histories  
245 (Oosterheld and Oyarzabal 2004; Bossuyt et al. 2005; Graff et al. 2007) and when unpalatable  
246 plants are recent invaders (Callaway et al. 2000, 2005). In habitats with a short history of  
247 grazing, most established plant species or genotypes have not evolved the ability to tolerate,  
248 resist, or avoid grazing and are thus very palatable. Therefore, protective effects by unpalatable  
249 plants are likely to greatly facilitate the performance of grazing-sensitive neighbor plants. In  
250 contrast, long grazing histories select for plant species or genotypes that can survive and  
251 reproduce under grazing pressures, and consequently the differences in grazing vulnerability

252 among plant species may be reduced. Rebollo et al. (2002) also suggested that in grasslands  
253 with a long evolutionary history of grazing, palatable species can independently tolerate or  
254 avoid grazing, rather than depending on the protective effects of unpalatable species. Our  
255 previous study examined the relative importance of positive and negative effects of *Urtica* on  
256 the dwarf-phenotype of *Persicaria* using exclosures and experimental removal of *Urtica*, and  
257 revealed that facilitative effects of *Urtica* were limited under natural conditions in Nara Park  
258 (Suzuki and Suzuki 2011a). Recently, we more rigorously analyzed the effects of *Urtica*  
259 considering distances between *Urtica* and *Persicaria* individuals, and revealed that the relative  
260 importance of positive and negative effects of *Urtica* on *Persicaria* varies temporally and  
261 spatially around a single *Urtica* plant (Suzuki and Suzuki 2011b). From the results of these  
262 previous studies and the present research, we conclude that the effects of *Urtica* on the  
263 dwarf-phenotype of *Persicaria* vary from facilitative to competitive under natural conditions,  
264 but the facilitative effects of *Urtica* on the dwarf-phenotype of *Persicaria* are generally limited.

265 Interactions among plants can lead to co-evolution and evolution of plant traits  
266 (reviewed in Thorpe et al. 2011). The evolutionary origins and maintenance of facilitation,  
267 however, have yet to be explored. Brooker et al. (2008) suggested that an evolutionary focus is  
268 absent from almost all recent work in the field of facilitation. Bronstein (2009) pointed out that  
269 the lack of evolutionary knowledge on facilitation is likely due to such topics having only  
270 recently been explored, and stressed that further understanding of the evolutionary origins and  
271 maintenance of facilitation must await studies that take an explicitly reciprocal approach that  
272 focuses on both the traits of facilitators and facilitated or beneficiary species and the genetic  
273 basis of their traits. Facilitation has the potential to promote the adaptation of beneficiary  
274 species to their facilitative neighbors (Ehlers and Thompson 2004; Thorpe et al. 2011).

275 However, facilitative traits may have costs for facilitators by helping their competitors.  
276 Michalet et al. (2011) found genetically based variation in facilitation of *Geum rossii*, and  
277 revealed that the facilitative phenotype suffered more competitive pressure from beneficiary  
278 species than the less facilitative phenotype. On the other hand, the results of the present study  
279 suggest that adaptation of palatable species (i.e., beneficiary species) to long-term grazing by  
280 developing “self-defensive” traits can lead to the dissolution of facilitative interactions between  
281 facilitators and beneficiary species. Together with the findings of these previous studies, the  
282 results of the present study suggest that facilitative interactions between species are not  
283 maintained during a long evolutionary period when phenotypic evolution of the facilitator and  
284 beneficiary species occurs.

285 Our results suggest that the values of each morphological trait tended to increase  
286 consistently from May to September, but decreased at the last census in October (Fig. 2). The  
287 reduced sizes in October may have been due to larger individuals being more likely to be grazed  
288 by deer than smaller plants at this time. Indeed, two individuals of the Heijo population had  
289 grown up over the nettle canopy in September, but they died in October due to deer grazing.  
290 Another possibility is that *Persicaria* plants had already begun to wither by October, which is  
291 the end of their growing season. Despite the seasonal variation in our results, we were able to  
292 identify the net effects of *Urtica* on two phenotypes of *Persicaria* by applying generalized  
293 linear mixed-effects models, which can include seasonal variation as a random factor. We also  
294 found increased numbers of leaves and shoots in plants located outside the *Urtica* canopy,  
295 which were likely a result of increased branching due to the release from apical dominance after  
296 suffering grazing damage. Our previous cultivation experiment using dwarf- and  
297 large-phenotypes of *Persicaria* also demonstrated that the number of leaves and shoots was

298 increased for artificially clipped individuals compared to intact individuals irrespective of plant  
299 phenotype, and the degree of plasticity in response to clipping was similar among populations  
300 (Suzuki 2008).

301 In conclusion, we demonstrated that the morphological differences of palatable plants  
302 shift the interactions between large unpalatable plants and the palatable plants from facilitative  
303 to competitive in a grazed community. Although previous studies have demonstrated that  
304 facilitative interactions play an important role in maintaining the biodiversity of plant  
305 communities, such conservation effects of facilitation may be weakened in plant communities  
306 with a long history of grazing.

307

308

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415

416 **FIGURE LEGENDS**

417 **Fig. 1** Seasonal change in the number of surviving plants for two populations (*triangle*, Nara  
418 population with dwarf phenotype; *circle*, Heijo population with large phenotype) in two  
419 positions [outside (*open*) and inside (*closed*) the *Urtica* canopy] during the experimental period  
420 after transplanting at the Nara Park site.

421  
422 **Fig. 2** Seasonal change in plant height (a), number of leaves (b), number of shoots (c), and  
423 plant size index (d) for surviving plants of two populations (*triangle*, Nara population with  
424 dwarf phenotype; *circle*, Heijo population with large phenotype) in two positions [outside  
425 (*open*) and inside (*closed*) the *Urtica* canopy] during the experimental period after transplanting  
426 at the Nara Park site (Mean  $\pm$  SE).

427  
428 **Fig. 3** Number of flowers in October and lifetime fitness for surviving plants of two  
429 populations (*open circle*, Nara population with dwarf phenotype; *closed circle*, Heijo  
430 population with large phenotype) in two positions at the Nara Park site (Mean  $\pm$  SE).

431

432

1 **Table 1** Results of the Cox proportional hazards regression model testing the effects of  
 2 population and distance on plant survival. The bold value indicates statistically significant.

	Coefficient	SE	<i>z</i>	<i>P</i>
Population (P)	-0.62	0.33	-1.87	0.061
Distance (D)	-0.25	0.30	-0.83	0.410
P × D	0.90	0.44	2.04	<b>0.041</b>

3

4

1 **Table 2** Results of the generalized linear mixed-effects model testing the effects of population  
 2 and distance on plant traits during a study period. The bold values indicate statistically  
 3 significant.

	Height		Number of leaves		Number of shoots		Plant size	
	Estimate	<i>P</i>	Estimate	<i>P</i>	Estimate	<i>P</i>	Estimate	<i>P</i>
Intercept	1.35	<b>&lt;0.0001</b>	2.35	<b>&lt;0.0001</b>	0.83	<b>0.003</b>	2.93	<b>&lt;0.0001</b>
Population (P)	-0.49	<b>&lt;0.0001</b>	0.35	<b>0.012</b>	0.37	<b>0.022</b>	-0.58	<b>0.001</b>
Distance (D)	0.07	0.505	-0.25	0.069	-0.36	<b>0.041</b>	-0.22	0.213
P × D	-0.25	0.074	-0.43	<b>0.033</b>	-0.28	0.247	-0.39	0.120

4

5

1 **Table 3** Results of the generalized linear mixed-effects model testing the effects of population  
 2 and distance on the number of flowers per individual and lifetime fitness in October. The bold  
 3 values indicate statistically significant.

	Number of flowers		Lifetime fitness	
	Estimate	<i>P</i>	Estimate	<i>P</i>
Intercept	2.09	<b>0.004</b>	-0.3211	0.785
Population (P)	0.52	0.271	1.5712	<b>&lt;0.0001</b>
Distance (D)	0.43	0.609	-0.5082	<b>&lt;0.0001</b>
P × D	-1.79	<b>0.031</b>	-2.1308	<b>&lt;0.0001</b>

4 The lifetime fitness of each plant that was estimated as the number of flowers produced by the  
 5 plant multiplied by the survival rate of plants in the treatment to which the plant belonged.

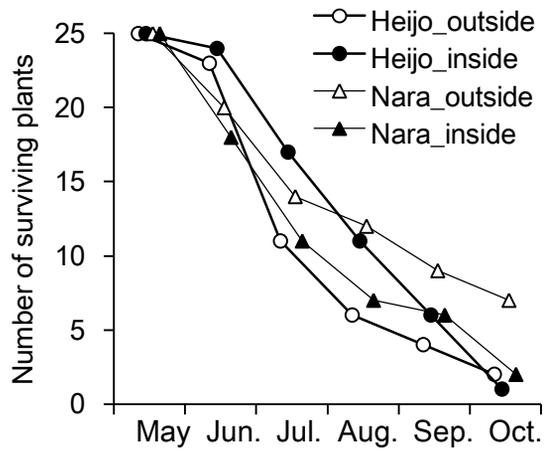


Fig. 1

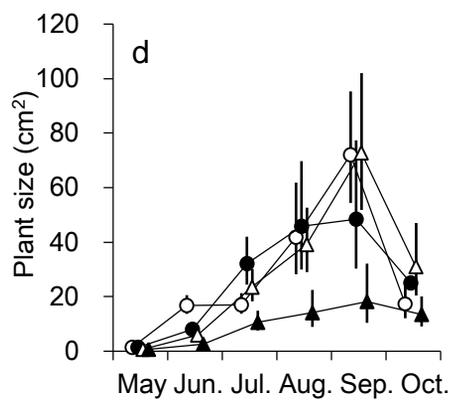
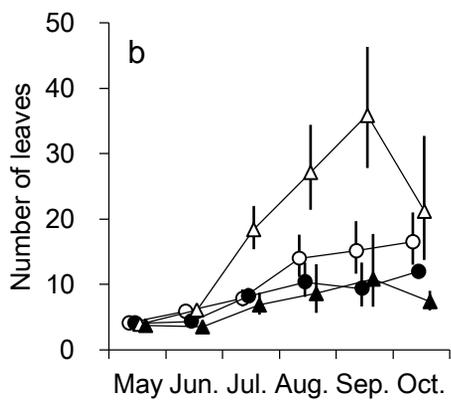
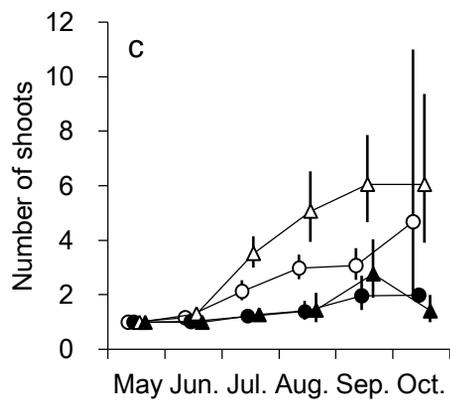
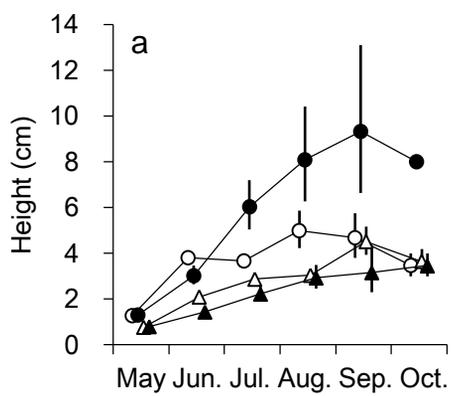


Fig. 2

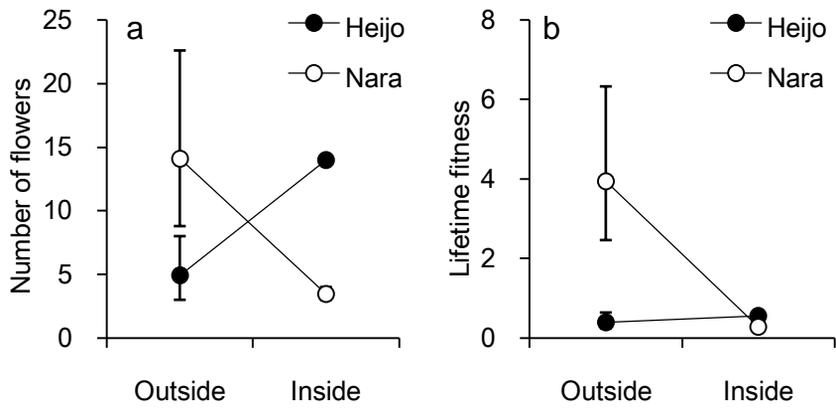


Fig. 3