TITLE:

Selection on flowering phenology mediated by seed predators and dependent on the community context

Butterfly seed predators and ants as selective agents for flowering phenology

Butterfly seed predators and their second hosts as selective agents for flowering phenology

Selection on flowering phenology: the role of (butterfly) seed predators and their second hosts

Selection on flowering phenology mediated by (butterfly) seed predators and their second hosts

Shifts in selection on flowering phenology mediated by (butterfly) seed predators and their second hosts

Context-dependent seed predation shifts direction of selection on flowering phenology

Predator-mediated and context-dependent (shifts in) selection on flowering phenology

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INTRODUCTION

Timing of reproduction in plants affects their fitness by influencing interactions both with the physical environment and with other organisms. Both pollinators (mutualists) and predispersal seed predators (antagonists) can exert selection on flowering phenology (Elzinga *et al.*, 2007) and this leads to different and sometimes opposing selective forces (Brody, 1997; Strauss & Irwin, 2004). Phenotypic selection on floral traits by pollinators is positively related to the degree of pollen limitation (Totland, 2001), and pollinator preference for certain floral traits does not always translate into higher fitness. However, preference of a phenotype by seed predators most likely decreases the fitness of that particular phenotype (Parachnowitsch & Caruso, 2008). Nowadays, there is weak empirical support for pollinator-mediated selection on flowering time (Ehrlén, 2015), but growing evidence for predispersal seed predators affecting selection on plant traits (Kolb *et al.*, 2007). The latter have been shown to exert phenotypic selection on flowering time (Ehrlén & Münzbergová, 2009) and in other reproductive traits as flower number (Leimu *et al.*, 2002) or inflorescence height (Kolb & Ehrlén, 2010). Plant phenology may be correlated with other traits also subject to natural selection (Lande & Arnold, 1983). Earlier flowering plants usually have more available resources and thus are able to grow higher and produce more flowers than later-flowering plants (Ison & Wagenius, 2014), causing a fitness decrease over the season due to an indirect effect of phenology. Selection on these correlated traits might act indirectly on flowering phenology (Kingsolver & Diamond, 2011).

Spatiotemporal variation in species interactions and in selection intensity on a given trait may result in selection mosaics (Thompson, 2005), and lead to different coevolutionary trajectories of plants and their animal interactors in different populations. For example, the relationship between plant reproductive traits and predispersal seed predation may vary among populations and years (Kolb *et al.*, 2007), and can contribute to the existence of geographic mosaics of coevolution between plants and their seed predators (Benkman, 1999; Rey *et al.*, 2006). Seed predators might thus vary in their importance as selection agents, due to variations in their abundance, and therefore in interaction intensity (predation rates), and due to variations in the direction and strength of predator trait preferences.

Both interaction intensities and preferences of seed predators might in turn be related to the environmental context. Both the physical environment where the interaction occurs (Kolb & Ehrlén, 2010; König *et al.*, 2014, 2015; von Euler *et al.*, 2014) and other community members (i.e. the community context, Brandt & Foitzik, 2004; Siepielski & Benkman, 2007) might influence interactions. In the simplest case of effects of community context, the outcome of an interaction between a pair of species (e.g. a plant and its seed predator) depends on the interaction with a third species present in the community (Strauss & Irwin, 2004).

Large Blue butterflies (*Maculinea* spp.) are predispersal seed predators of specific host plants during their first larval instars. However, *Maculinea* larvae need a second host to complete their development, and they are parasites of ant nests (*Myrmica* spp.) during later instars (Als *et al.*, 2004). The community context in terms of the abundance of the second host might thus indirectly affect phenotypic selection on host plant traits if it influences butterfly abundance and predation intensity.

In this study, we examined how phenotypic selection on flowering time in the perennial herb *Gentiana pneumonanthe* is mediated by the interaction with its specialist predispersal seed predator, the butterfly *Maculinea alcon*, and how this interaction depends on the community context in terms of the abundance of the second host. We addressed two main questions: 1) Is phenotypic selection on flowering phenology in *G. pneumonanthe* mediated by the interaction with its seed predator (*M. alcon*)? and 2) Are *M. alcon* presence and seed predation intensity related to the community context in terms of the abundance of their second host (*Myrmica* ants)?

MATERIALS AND METHODS

**Study system**

The marsh gentian (*Gentiana pneumonanthe* L.) is a rare, long-lived perennial herb that may reach 45 cm in height, overwintering as a rosette of short shoots with small leaves (Simmonds, 1946). Plants can have one to many shoots and produce deep blue flowers pollinated by bumblebees (although the species is entirely self-compatible). It flowers in July and August in SW Sweden. Fruits are capsules containing a high number of minute, wind-dispersed seeds. The species inhabits open habitats as wet heathlands and grasslands. Although it appears in some other places in Europe, it is red-listed as vulnerable in Sweden. *G. pneumonanthe* is the primary host of the Alcon Blue butterfly (*Maculinea alcon*), a specialist predispersal seed predator which lays its eggs on young gentian buds during its flight period (July and August, Appelqvist *et al.*, 2007). The caterpillars consume a large fraction of developing seeds in some populations. Fourth-instar caterpillars drop to the ground and are then picked up by their second host: *Myrmica* ants (probably *M. ruginodis* in our study area, Appelqvist *et al.*, 2007). Caterpillars mimic the surface chemistry of the ant brood (Nash *et al.*, 2008), and this makes ants carry them to their nest, where they spend the rest of their larval period as parasites (Mouquet *et al.*, 2005). Contrary to the majority of *Maculinea* species, which prey on ant brood, *M. alcon* is a “cuckoo” species (Als *et al.*, 2004), whose larvae are fed primarily on regurgitations from ant workers, trophic eggs and prey items.

**Data collection**

We collected data on reproductive traits, plant fitness and interaction intensity in 100 marked individuals in each of the study populations during 2010 and 2011. For each individual, we selected one focal shoot as the one having median length. All measures were carried out on this focal shoot. We measured shoot height, phenological state, number of flowers and number of *M. alcon* eggs in late July – early August. Phenology of each individual, in terms of the developmental stages of flowers, was estimated by counting the number of buds and flowers in each of six developmental stages: a) small bud (the sepals covering the bud completely), b) medium-sized bud (from the time the bud begins to protrude outside the sepals until it grows over the sepals) c) large bud (the bud becomes elongated and rather high), d) not fully blooming flower (from the time the bud starts changing into blue until it becomes a solid blue, tightly twisted bud) e) blooming flower (from the time the bud starts to open, when the petals start to spin up, to a fully opened flower) and f) wilted flower (merged and started to wither). We extracted two different measures of phenology. First, a phenological index (PI) was calculated for each individual as PI = (1a + 2b + 3c + 4d + 5e + 6f) / nfl, with nfl = total number of flowers and buds. Second, the state of the most advanced bud in each shoot was recorded (a-f).

Populations were visited until all fruits had matured (until mid-October), and the maximum number of eggs and the final number of intact fruits (i.e. not damaged by the butterfly) were assessed and used as estimates of interaction intensity and plant fitness, respectively. In 2011, no information on final fruit production was available for 4 of the populations.

Ant abundance was measured in each of the 20 study populations using ant counts. In 2010, a 20-m transect was established through each population, and a sugar cube was placed every meter (i.e. 20 sugar cubes per population). In 2011, we used a 40-m transect with 40 sugar cubes in each population. The number of *Myrmica* sp. ants in each of the cubes was counted after 30 minutes. We then calculated the average number of ants per sugar cube in each population and year, and used the highest of both yearly values (in order to compensate for underestimations due to e.g. adverse climatic conditions) as a measure of ant abundance for each population.

**Statistical analyses**

To evaluate if there was phenotypic selection on flowering phenology, we performed selection gradient analyses (Lande & Arnold, 1983) by regressing relative fitness on standardized estimates of phenology, together with other reproductive traits (flower number and shoot height). Within each population, absolute fitness (number of intact fruits) was relativized to have a mean of one (by diving it by the mean number of intact fruits), and reproductive traits (phenology, flower number and shoot height) were standardized to have a mean of zero and a standard deviation of one (by subtracting the mean and dividing by the standard deviation). To test for differences in phenotypic selection gradients among populations, we included also interactions between standardized reproductive traits and population. The main effect of population was not included because fitness was relativized within populations prior to analysis. We fitted a linear model with Type II sums of squares. We tested for directional selection (linear terms), but also for correlational selection (examining the effects of interaction terms in a model also including the linear terms) and non-linear selection (examining the effect of quadratic terms in a model also including the linear terms). Results for PI and state of the most advanced bud were very similar in all models. Below, we present only results for the state of the most advanced bud, hereafter referred to as “phenology” variable (higher values indicating earlier flowering).

To test if the observed among population-variation in linear selection on reproductive traits (see Results) was mediated by the interaction with *M. alcon*, we created a dummy variable “Predation”, coded as 0 (in populations without *M. alcon*) and 1 (in populations with *M. alcon*), and constructed a linear hierarchical mixed model (Type II sums of squares) for each year, with relative fitness as response variable. Predictors included standardized reproductive traits, their interactions with population (as random effects) and with predation (as fixed effects). Trait x population and trait x predation interactions were only included if the trait x population interaction was significant. We did not include an individual intercept by population because fitness was relativized within populations prior to analysis. To restrict model complexity and because other effects showed to be less important (see Results), we consider only linear effects in these models.

Within each population, we estimated linear selection gradients (’s) from multiple regressions of relative fitness on standardized reproductive traits, and we compared linear selection gradients for phenology between populations with and without predator using one-way ANOVAs.

To investigate among-population variation in predator preferences, i.e. the relationships between the occurrence and intensity of predation and reproductive traits, we regressed both the probability of attack by *M. alcon* (using a dummy variable “attack”, coded as 0 when number of eggs = 0, and 1 when number of eggs > 0) and the number of eggs on reproductive traits, population and their interactions. We excluded data from population D in 2010 from these analyses because predation intensity was extremely low (only 1 plant with 2 eggs).

We also examined the causal effects of phenology and other reproductive traits on fitness using path analyses (Grace, 2006) in the subset of populations where the predator was present (data from population D in 2010 was excluded also). We included both direct and indirect effects of traits on fitness, the latter mediated by the probability of predator attack or interaction intensity. To test for spatial variation in selective scenarios among populations, we used multigroup analysis (Grace, 2006). Afterwards, we fitted the path models in each population separately because they differed statistically. Details and results of this additional analysis are given in Appendix S2.

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Finally, we examined if abundance of the second host of *M. alcon* influeces phenotypic selection by increasing the probability of predator presence or the intensity of the interaction. For this, we performed a) a logistic regression of butterfly presence on log-transformed ant abundance, using the 20 study populations, and b) per-year linear regressions of the mean number of eggs per plant and the proportion of plants with *M. alcon* eggs on log-transformed ant-abundance, using the 11 populations where the butterfly was present.

Analyses were conducted in R 3.1.2 (R Core Team, 2014), with the exception of the path analyses shown on Appendix S2, which were conducted in Amos 16.0 (Arbukle, 2007).

RESULTS

We found evidence for linear (directional) selection on flowering phenology, flower number and shoot height in both study years (Table 1A). Linear selection on flowering phenology and flower number varied among populations in both study years. Linear selection on shoot height did not differ among populations in 2010, with relative fitness increasing in plants with higher shoots, but varied among populations in 2011. We also found evidence of correlational selection. Some trait interactions showed significant effects (Table 1B), although they differed among populations. In both study years, we found a significant quadratic effect of flower number on fitness that differed among populations (model C in Table 1).

Phenotypic selection on flowering phenology differed between populations where the predator was absent and populations where it was present (Table 2, Appendix S3). In populations where the predator was absent, there was selection for earlier flowering (mean ± 95% CI of selection gradients, 2010: 0.22 ± 0.15, 2011: 0.30 ± 0.17), while in populations where the predator was present there was selection for later flowering (mean ± 95% CI of selection gradients, 2010: -0.19 ± 0.15, 2011: -0.10 ± 0.11, Fig. 1).

In both study years, early flowering increased the probability of being attacked by the predator, and this effect was constant between populations (Table 3, Appendix S4). In 2010, there were differences between populations in the relationship between probability of attack and flower number, while in 2011 the predator consistently preferred plants with higher number of flowers in all populations. In 2011, there were differences between populations in the relationship between probability of attack and shoot height. There were also differences between populations in the effects of phenology and flower number on predation intensity (number of eggs, Table 3, Appendix S4) in both years studied.

Multigroup analyses revealed significant among-population variation in selective scenarios (Tables S2.1-2). Although there was a high variation between populations and years, models fit for each population (Tables S2.3-5) show that indirect effects of phenology on fitness (i.e. mediated by the seed predator preference for early-flowering plants, which in general reduced their fruit production) were more commonly significant than direct effects.

The probability of presence of *M. alcon* was higher in populations with higher abundance of *Myrmica* ants (Fig. 2A). Although there were populations with high ant abundance where *M. alcon* was absent, in populations where it was present ant abundance was generally high. Neither the mean number of eggs per plant (Fig. 2B) nor the proportion of plants with *M. alcon* eggs (Fig. 2C) was related to ant abundance in any of the two years.

DISCUSSION

In this study, we have shown that the interaction between *G. pneumonanthe* and its seed predator *M. alcon* is responsible for phenotypic selection on flowering phenology. *M. alcon* selects for later flowering in its host plant, as earlier flowering increases the probability of predator attack and the intensity of predation. Phenotypic selection on flowering phenology is dependent on the community context, as presence of the predator (although not predation intensity) is related to ant abundance.

Our finding that there is phenotypic selection for later flowering phenology mediated by the interaction with a butterfly seed predator agrees with previous studies demonstrating predator-mediated selection on phenology (Pilson, 2000; Parachnowitsch & Caruso, 2008). However, other studies (see review by Kolb *et al.*, 2007) have reported predators selecting for both early and late flowering. . Therefore, plants flowering late have a higher fitness in presence of the predator because they are less prone to be attacked.

Our results also show that in the absence of the predator, phenotypic selection favored early flowering, being consistent with the direction of the general trend shown by Munguía-Rosas *et al.* (2011). This could be due to early flowering directly increasing fitness (e.g. by favoring outcrossing, Munguía-Rosas *et al.*, 2011), but also to indirect selection on other correlated reproductive traits (e.g. early-flowering plants might have more resources and produce in general higher shoots and more flowers), or to environmental covariance (i.e. phenology and fitness being both influenced by the environment, Ehrlén, 2015). In this case, the effect of phenology on fitness remains significant even after including two traits (flower number and shoot height) representing plant resources as covariates in our models, indicating that early flowering directly increases fitness in the absence of the predator. Therefore, we have demonstrated that the direction of selection on phenology differs among populations and that this variation can be explained by *M. alcon* presence: the predator is responsible for shifting selection from early to late flowering. In a recent meta-analysis (Siepielski *et al.*, 2013), selection was reported to vary mainly in strength, but less in direction among populations. We have found evidence of variation both in direction (i.e. favoring early or late flowering depending on absence or presence of the predator) and in strength (cf. linear selection gradients in Appendix S3). If predator distribution is constant in time, and there is sufficient genetic variance in flowering time to allow a genotypic selection response (Putterill *et al.*, 2004), this divergent selection could also lead to local adaptation (Thompson, 2005) and result in genetic differences in flowering phenology among populations. Besides, the differences in selection strength could lead to populations experiencing stronger selection adapting more quickly than populations experiencing weak selection (Siepielski *et al.*, 2013). Our data do not show significant differences in flowering phenology among populations where the predator is present and absent (results not shown). However, common garden experiments are ongoing in order to assess if these populations differ in flowering phenology when environmental variation is removed.

We also found evidence for phenotypic selection on other plant traits. In 2010, plants with higher shoots were positively selected in all populations, while in 2011, there were differences between populations in selection for shoot height that were mediated by seed predation. Butterflies might prefer to lay eggs on tall shoots, as this indicates better plant quality and hence more resources available for larval development (Czekes *et al.*, 2014; Wynhoff *et al.*, 2014). However, this was only the case in some populations and years in our study system (Fig. S4.3). Phenotypic selection on flower number varied among populations, but these differences were not mediated by predation, and could be explained by variation in the strength of the relationship between flower and fruit production (due to e.g. variation in pollination success or in resources available for fruit set, Zimmerman & Aide, 1989).consistently The visibility of food plants for butterflies may be determined by both its height and the size of its floral display (Nowicki *et al.*, 2005). However, in our study system, the first factor is not very important, and the importance of the second varies between years.

Selection on flowering phenology in *G. pneumonanthe* depends on the presence of the predator, and thus is also indirectly determined by the factors that determine predator presence. We have shown that the community context in terms of the abundance of the second host is a key factor for predator presence, and thus it will indirectly condition phenotypic selection on flowering phenology. Previous studies have shown that the community context can affect the likelihood or intensity of plant-animal interactions (Strauss & Irwin, 2004), as selection on plant traits by either mutualists or antagonists can be influenced by other community members. For example, nectar robbers can affect selection by pollinators (Irwin, 2006), and plant neighborhood may alter selection by insect herbivores (Agrawal *et al.*, 2006). In our study system, the presence of *M. alcon* is more probable in sites with a high abundance of *Myrmica* ants, as the butterfly needs them to complete its life cycle. Therefore, the plant-seed predator interaction can only take place and mediate phenotypic selection in these sites. However, in populations where the butterfly is present, the intensity of seed predation on *G. pneumonanthe* is not related to ant abundance. This suggests that the second host is crucial for antagonist presence, but does not determine its abundance, at least at the population scale. Further studies relating predation to ant abundance near individual plants would clarify if community context determines interaction intensity at an individual scale, as the evidence for ant-related oviposition patterns is nowadays controversial (van Dyck *et al.*, 2000; Nowicki *et al.*, 2005; Fürst & Nash, 2010; Wynhoff *et al.*, 2014).

In summary, we have shown that biotic interactions can shift the direction of phenotypic selection, and that community context can indirectly explain the variation in selection among populations. In our study system, variation in selection on flowering phenology is determined by a seed predator, whose presence is conditioned by the abundance of its second host. Thus, both antagonist interactors and the community context may act respectively as direct and indirect selective agents that can determine evolution on flowering time. Longer studies that also investigate the heritability of phenological traits are needed to assess if this shift in selection is maintained in time and could result in genetic differences in flowering phenology among populations.

ACKNOWLEDGEMENTS

We thank Susanne Govella, Anna Herrström and Jessica Oremus for field data collection and Johan P. Dahlgren for statistical advice. We acknowledge funding from the Swedish Research Council to JE and from the “Clarín” postdoctoral program (FICYT, Gobierno del Principado de Asturias, Spain, and Marie Curie-Cofund Actions, EU) to AV.

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TABLES

Table 1: Effects of standardized traits (flowering phenology, flower number, and shoot height), population and their interactions on relative fitness (number of intact fruits) of *G. pneumonanthe* in 2010 (N = 2000 plants in N =20 populations) and 2011 (N = 1598 plants in N = 16 populations). Results are from linear models with Type II sums of squares, including: A) only linear effects, B) linear effects and interactions and C) linear and quadratic effects. Estimates are given for significant main effects where the Population x trait interaction is not significant.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Source of variation | |  | 2010 |  |  |  | 2011 |
| df | F | Estim |  | df | F |
| A) Linear terms | |  |  |  |  |  |  |
|  | Phenology (early flowering) | 1 | 0.02 |  |  | 1 | 0.50 |
|  | Flower number | 1 | 183.73\*\*\* |  |  | 1 | 50.81\*\*\* |
|  | Shoot height | 1 | 7.18 \*\* | 0.128 |  | 1 | 0.47 |
|  | Population x Phenology | 19 | 3.61\*\*\* |  |  | 15 | 1.98\* |
|  | Population x Flower number | 19 | 3.88\*\*\* |  |  | 15 | 2.90\*\*\* |
|  | Population x Shoot height | 19 | 1.19 |  |  | 15 | 2.35\*\* |
|  |  |  |  |  |  |  |  |
| B) Interaction terms | |  |  |  |  |  |  |
|  | Phenology x Flower number | 1 | 1.78 |  |  | 1 | 0.08 |
|  | Phenology x Shoot height | 1 | 3.21 |  |  | 1 | 2.16 |
|  | Flower number x Shoot height | 1 | 9.59\*\* |  |  | 1 | 4.57\* |
|  | Population x Phenology x Flower number | 19 | 2.66\*\*\* |  |  | 15 | 1.61 |
|  | Population x Phenology x Shoot height | 19 | 1.02 |  |  | 15 | 1.31 |
|  | Population x Flower number x Shoot height | 19 | 1.82\* |  |  | 15 | 2.37\*\* |
|  |  |  |  |  |  |  |  |
| C) Quadratic terms | |  |  |  |  |  |  |
|  | Phenology 2 | 1 | 2.47 |  |  | 1 | 0.13 |
|  | Flower number 2 | 1 | 4.38\* |  |  | 1 | 2.02 |
|  | Shoot height 2 | 1 | 0.03 |  |  | 1 | 1.50 |
|  | Population x Phenology 2 | 19 | 1.40 |  |  | 15 | 0.94 |
|  | Population x Flower number 2 | 19 | 2.40\*\*\* |  |  | 15 | 3.38\*\*\* |
|  | Population x Shoot height 2 | 19 | 1.22 |  |  | 15 | 1.80\* |

\* p < 0.05; \*\* p < 0.01; \*\*\* p < 0.001

Table 2: Effects of standardized traits (flowering phenology, flower number, and shoot height), predation and their interactions on relative fitness (number of intact fruits) of *G. pneumonanthe* in 2010 (N = 2000 plants in N =20 populations) and 2011 (N = 1598 plants in N = 16 populations). Results are from linear hierarchical mixed models with Type II sums of squares (Wald 2 values are shown). Trait x population interactions that were significant in Model A in Table 1 are included here as random effects. Boldface indicates significance.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Source of variation | 2010 | |  | 2011 | |
| df | 2 |  | df | 2 |
| Phenology (early flowering) | 1 | 0.00 |  | 1 | 0.86 |
| Flower number | 1 | 53.85\*\*\* |  | 1 | 27.82\*\*\* |
| Shoot height | 1 | 7.82\*\* |  | 1 | 0.21 |
| Predation | 1 | 0.11 |  | 1 | 0.00 |
| Predation x Phenology | 1 | 14.72\*\*\* |  | 1 | 14.95\*\*\* |
| Predation x Flower number | 1 | 0.01 |  | 1 | 3.69 |
| Predation x Shoot height | 1 | - |  | 1 | 5.69\* |

\* p < 0.05; \*\* p < 0.01; \*\*\* p < 0.001

Table 3: Effects of population, phenology, flower number and shoot height of *G. pneumonanthe* on attack by *M. alcon* and number of eggs in 10 populations in 2010 (N = 1000 plants) and 11 populations in 2011 (N = 1099 plants). 2 values are shown for logistic regressions, and F values for linear regressions. Estimates (from a model without interaction terms) are given for significant main effects where the interaction with population is not significant Boldface indicates significance.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Source of variation | Response: attack | | | | | | |  | Response: number of eggs | | | | |
| 2010 | | |  | 2011 | | |  | 2010 | |  | 2011 | |
| df | 2 | Estim |  | df | 2 | Estim |  | df | F |  | df | F |
| Phenology (early flowering) | 1 | 39.32\*\*\* | 0.420 |  | 1 | 39.00\*\*\* | 0.545 |  | 1 | 79.57\*\*\* |  | 1 | 46.87\*\*\* |
| Flower number | 1 | 27.69\*\*\* |  |  | 1 | 81.15\*\*\* | 0.794 |  | 1 | 75.43\*\*\* |  | 1 | 240.98\*\*\* |
| Shoot height | 1 | 1.93 |  |  | 1 | 0.04 |  |  | 1 | 0.01 |  | 1 | 0.03 |
| Population | 9 | 181.74\*\*\* |  |  | 10 | 138.77\*\*\* |  |  | 9 | 29.09\*\*\* |  | 10 | 46.00\*\*\* |
| Population x Phenology | 9 | 8.62 |  |  | 10 | 16.65 |  |  | 9 | 9.91\*\*\* |  | 10 | 5.55\*\*\* |
| Population x Flower number | 9 | 29.67\*\*\* |  |  | 10 | 14.11 |  |  | 9 | 8.77\*\*\* |  | 10 | 33.61\*\*\* |
| Population x Shoot height | 9 | 9.98 |  |  | 10 | 18.38\* |  |  | 9 | 0.52 |  | 10 | 0.72 |

\* p < 0.05; \*\* p < 0.01; \*\*\* p < 0.001

FIGURE LEGENDS

Figure 1: Differences in linear selection gradients for phenology between populations where the predator was present (*M. alcon* =1, N = 11) and absent (*M. alcon* = 0, N = 9) in 2010 and 2011. P-values from one-way ANOVAs are shown.

Figure 2: Effects of abundance of the second host (log-transformed maximum *Myrmica* abundance for both years) on A) *M. alcon* presence (p from logistic regression fit is shown, N = 20 populations), B) Mean number of eggs per plant, and C) Proportion of plants with *M. alcon* eggs. In B) and C), N = 11 populations where the predator was present, symbol color indicates the study year for interaction intensity measures (black = 2010 and grey = 2011).

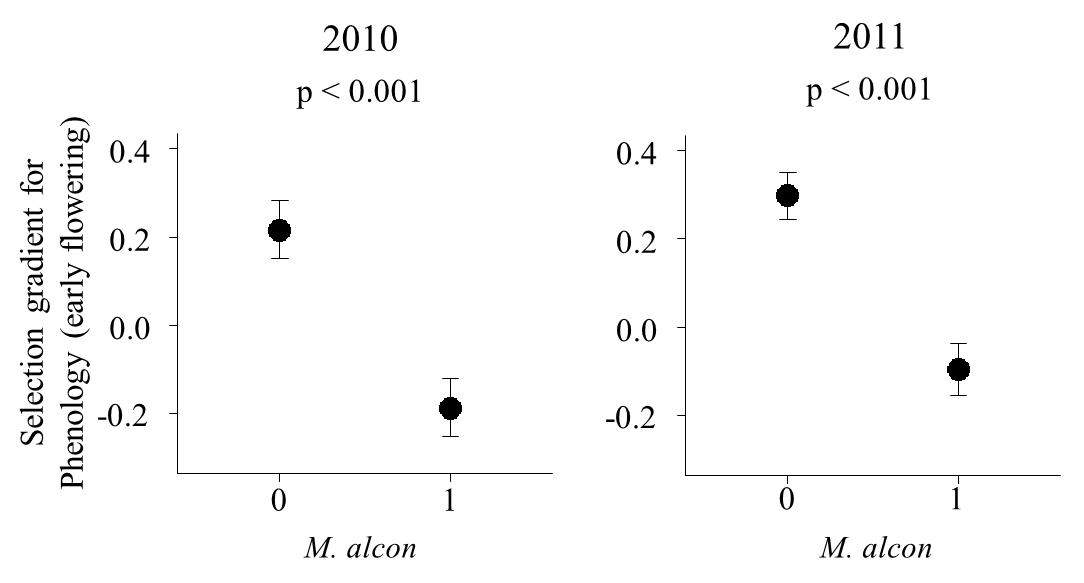
Figure 1

Figure 2

