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Local adaptation and ecological genetics of host-plant specialization in a leaf beetle

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The tendency of insect species to evolve specialization to one or a few plant species is probably a major reason for the remarkable diversity of herbivorous insects. The suggested explanations for this general trend toward specialization include a range of evolutionary mechanisms, whose relative importance is debated. Here we address two potentially important mechanisms: (i) how variation in the geographic distribution of host use may lead to the evolution of local adaptation and specialization; (ii) how selection for specialization may lead to the evolution of trade-offs in performance between different hosts. We performed a quantitative genetic experiment of larval performance in three different populations of the alpine leaf beetle *Oreina elongata* reared on two of its main host plants. Due to differences in host availability, each population represents a distinctly different selective regime in terms of host use including selection for specialization on one or the other host as well as selection for utilizing both hosts during the larval stage.

The results suggest that selection for specialization has lead to some degree of local adaptations in host use: both single-host population had higher larval growth rate on their respective native host plant genus, while there was no difference between plant treatments in the two-host population. However, differences between host plant treatments within populations were generally small and the degree of local adaptation in performance traits seems to be relatively limited. Genetic correlations in performance traits between the hosts ranged from zero in the two-host population to significantly positive in the single-host populations. This suggests that selection for specialization in single host populations typically also increased performance on the alternative host that is not naturally encountered. Moreover, the lack of a positive genetic correlation in the two host-population give support for the hypothesis that performance trade-offs between two host plants may typically evolve when a population have adapted to both these plants. We conclude that although there is selection for specialization in larval performance traits it seems as if the genetic architecture of these traits have limited the divergence between populations in relative performance on the two hosts.

The evolution and ecology of insect-host plant interactions is the subject of a large body of work (Ehrlich and Raven 1964, Jermy 1984, Futuyma and Moreno 1988, Thompson 1988a, 1994, Jaenike 1990, Futuyma 1991). Much of the effort has been devoted to explain the remarkable diversity of herbivorous insects, and the results suggest that one of the main explanations is the tendency of insect species to evolve specialization to

one or a few plant species (Futuyma and Moreno 1988, Jaenike 1990, Futuyma 1991, Thompson 1994). Questions about the potential causes of this evolutionary drive towards specialization have spurred discussion (Bernays and Graham 1988, Courtney 1988, Rausher 1988, Thomson 1988b). However, a consensus opinion appears to be emerging, which stresses that there is not one single explanation for the general evolutionary drive towards specialization but several (Futuyma 1983, 1991, Bernays and Graham 1988 and responses to that paper, Futuyma and Moreno 1988, Thompson 1988a, 1994, Jaenike 1990, Joshi and Thompson 1995, Janz and Nylin 1997, Bernays 1998).

A large number of the selective pressures that has been put forward to explain host plant specialization suggest that the evolutionary processes will be strongly dependent on geographic variation in insect-plant interactions (Thompson 1994). Indeed, geographic variation of species interactions seems to be the rule rather than the exception and it can, for example, be maintained by non-overlapping distributions of an insect species and all of its host plants, by spatial genetic variation in insects or hosts plants or geographically variable natural enemy fauna (Thompson 1994, Via 1994). To study situations where some level of local adaptation can be expected may therefore highlight evolutionary processes that are of relevance for the field of insect-host plant biology in general (Singer et al. 1994, Mopper 1996, 1998, Thomas and Singer 1998).

To what degree genetic categories (populations, demes etc) are locally adapted to their particular locality depends essentially on the relative rates of local selection and gene flow between genetic categories (Slatkin 1985, 1987, Barton and Whitlock 1997, Peterson and Denno 1998). However, the exact outcome of local selection on host plant utilization will also depend on the genetic architecture within the populations in question, i.e. is genetic variation for host use present? what is the nature of any genetic correlations between performance on different hosts? Furthermore, the process of local adaptation and specialization will not only depend on genetic architecture, it will also affect it (Rausher 1988, Thompson 1994, Joshi and Thompson 1995). In this study we aim to test some predictions of local adaptation and to investigate the genetic architecture of larval performance traits in the alpine leaf beetle *Oreina elongata* Suffrian (Coleoptera: Chrysomelidae). The spatial distribution of this beetle and its host plants, together with low rates of beetle dispersal, suggest that local adaptations in host use is likely to evolve.

We investigated larval growth performance on two host plant species (*Adenostyles alliariae* and *Cirsium spinosissimum*) of larvae from three different populations in the western Alps that represent three different categories of host plant use. The population at Col du Lautaret has no *C. spinosissimum* in its habitat and feeds almost exclusively on *Adenostyles glabra*, whereas the population at the Mattmark dam only has *C. spinosissimum* available and is exclusively found on this plant. Finally, the population at Col du Petit Saint Bernard has both *A. alliariae* and *C. spinosissimum* in its habitat and all life stages of *O. elongata* can be found on both host-species. The shortest distance between any of these populations is at least 200 km and they are separated by high altitude mountain ranges (2500 m–4000 m) making gene flow between the populations extremely unlikely.

The single-host populations (Mattmark – *A. glabra*, and Lautaret – *C. spinosissimum*) are only experiencing selection for performing well on their respective native hosts, while the two-host population (Petit St-Bernard – *A. alliariae* and *C. spinosissimum*) most likely experience selection on both plants. Earlier studies have shown that many individuals in this last population in fact feed on both plants during their larval period (Ballabeni et al. 2001a). Thus, we expected the single-host populations to be locally adapted for using their native host plant genus (*Adenostyles* and *Cirsium* respectively) and therefore to have higher larval performance on those plants compared to the alternative plant, while the two-host population should show a smaller difference in performance between host plants.

We furthermore hypothesised that directional selection should have reduced genetic variation for growth performance on the native plants of each population, while there may be more genetic variation for growth performance on the alternative plant in the two single-host populations. Hence, we expected the two single-host populations to have higher heritabilities of larval performance traits on the non-native plant species, while in the two-host population we did not expect any differences in heritabilities of larval performance between plant species.

Finally, we wanted to test if genetic correlations in performance traits on the different hosts may show differences between populations in line with a hypothesis proposed by Joshi and Thompson (1995), that suggests how trade-offs in growth performance between different host plants may evolve. The presence of such trade-offs is a commonly suggested explanation for host plant specialization in herbivorous insects, but despite a large number of studies investigating the matter there is little empirical evidence for the presence of negative genetic correlations in performance on different hosts (reviewed by Joshi and Thompson 1995). The authors stress that negative genetic correlations between hosts are most likely to arise if the insect population in question has been under selection on all host and has evolved to genetic equilibrium. They argue that in an insect population that uses two host species, only the alleles that provide negative pleiotropic effects between the hosts should remain variable and this should result in negative genetic correlations being present. On the

contrary, if only one of the two hosts species is established and the other is new within a given insect population, the alleles that affect fitness on the new host and those with between-host positive pleiotropic effects should still remain variable (have not gone to fixation yet) and they would reduce or hide any trade-off caused by alleles with negative pleiotropy (Joshi and Thompson, 1995). Our three populations correspond very well to the hypothesised testing situation described by Joshi and Thompson (1995) and provides and opportunity to evaluate their hypothesis. Consequently, we expected to find negative genetic correlations across hosts in our two-host population, while in the single-host populations these correlations would be more positive.

Materials and methods

Study organisms

Due to its alpine habitat the distribution of *O. elongata* populations is heterogeneous and field observations as well as mark-recapture studies strongly suggests that dispersal rates are very low. Despite many years of intensive studies there has been no observation of flying beetles (personal observations, Conconi, D. unpubl.) and it seems likely that *O. elongata* disperse mainly by walking.

Oreina elongata feeds and lays its eggs on three plant species that belong to two different tribes of the family Asteraceae: *Adenostyles alliariae* (Guoan), *A. glabra* (L.) (Senecioneae) and *Cirsium spinosissimum* (L.) (Carduaeae). In the Alps, the availability of these three hosts varies geographically and there are *O. elongata* populations that only have *Adenostyles*-species available in their habitats and others that only have *C. spinosissimum*, while others again have both an *Adenostyles*-species and *C. spinosissimum* present in their habitat. The two species in the genus *Adenostyles* are closely related and both contain pyrrolizidine alkaloids that are sequestered by adults and larvae of *O. elongata* to provide a chemical defence against natural enemies (Dobler et al. 1996). In the present investigation we treat these two plants as representing one host-type for *O. elongata*. The leaves of *C. spinosissimum* do not provide any sequestrable chemical defences but their dentate, hairy and spiny structure seems to give *O. elongata*'s eggs with some protection against natural enemies (Ballabeni et al. 2001b). Beetle populations that live in places where *C. spinosissimum* is the only host present rely on small amounts of self-synthesized cardenolides for chemical protection (Dobler and Rowell-Rahier 1994). Pyrrolizidine alkaloids seem to protect the beetles more efficiently, at least against generalist avian predators (Rowell-Rahier et al. 1995).

At Petit St-Bernard, *A. alliariae* and *C. spinosissimum* grow in patches of various sizes, which include either the one or the other plant species or both. In this population, *O. elongata* eggs are laid much more frequently, and have higher survival rates, on *C. spinosissimum* than on *A. alliariae* (Ballabeni et al. 2001a, b). On the other hand, larvae perform better when they feed on *A. alliariae* alone or on a mixture of *A. alliariae* and *C. spinosissimum* than when they feed on the latter plant alone (Ballabeni and Rahier 2000a).

Experimental design and procedure

We performed our study in a building located at Petit St-Bernard, about 500 m away from the local beetle population. We chose to do work in this location rather than in the laboratory at the University of Neuchatel because we needed fresh *A. alliariae* and *C. spinosissimum* to breed the larvae.

The experiment was organised as a quantitative genetics family-design with larval families from each of the three populations mentioned above. Larvae of a given family were produced from the eggs laid by a single mother collected in the field. Thus, the fathership was unknown and we had to work under the assumption of having full-sib families (Falconer 1989). A classic half-sib design with each male mated to several virgin females could not be used because the laboratory breeding of *O. elongata* bears low success rates and would necessitate about one year from egg to adult. Ten larvae of each family were tested for growth rate, developmental time and survival on *A. alliariae* and 10 on *C. spinosissimum*.

The experiment was performed with 24 families from Lautaret (département de Haut Alpes, France, altitude 2058 m), 25 from Mattmark (Canton Valais, Switzerland, altitude 2200 m) and 26 from Petit St-Bernard (at the border between the département de Savoie, France, and the region Vallée d'Aoste, Italy, altitude 2200 m). Mated, field collected females were individually kept in transparent, round plastic boxes of 90 mm diameter and 50 mm height, where they were allowed to lay eggs. The position of boxes was randomised on shelves in the laboratory. We collected the females from their host plants in early July 1999. All three populations were sampled within five days. We did not keep record of the plant species from which the Petit St-Bernard females were collected since the host on which females are found does not influence larval performance in this population (Ballabeni and Rahier 2000a). Each female was simultaneously fed with *A. alliariae* and *C. spinosissimum* during the egg laying period. Food plants were freshly collected in the field and renewed every 3 days.

The room temperature during the oviposition fluctuated between 7 and 17°C. This range lies within the limit of the natural summer temperature fluctuations at Petit St-Bernard and very likely at Lautaret and

Mattmark as well. The photoperiod followed the natural seasonal changes since the experiment was performed in a room with windows.

Oviposition was checked daily and each newly hatched larva was transferred into an individual petri dish of 30 mm diameter with a moist chalk bottom covered with a filter paper, to keep humidity. Each larva was reared during the whole experiment within its individual petri dish. Larvae were randomly assigned to either diet level according to the experimental design. During the whole experiment, we fed the larvae ad libitum with leaves collected in the same day in the field. We changed the food every 2 days. The petri dishes were randomised on shelves in the same room that was used for oviposition. Temperature and light conditions were thus the same as for the egg-laying females.

The larvae were checked daily for mortality and larval stage. We weighed each larvae on the hatching day and one day after the third moult and we noted the number of days from hatching to third moult. Larvae were not weighed on the exact day of their third moult because moulting is accompanied by large and inconsistent water losses which makes comparisons impossible, whereas weight differences are consistent one day after moult. We ended the experiment at third moult, e.g. at the beginning of the last larval instar, rather that at the pupal stage because *O. elongata*'s pupation rate is very low in the laboratory.

We calculated or recorded the following performance variables for each individual larva: daily growth rate (mg weight increase per day between hatching and 1 day after moult), development time (number of days from hatching to third moult), final weight (weight one day after third moult) and survival (surviving until third moult or not).

Data analysis

Since we had to analyse our data under the assumption of a full-sib design, our estimates of heritabilities and genetic correlations may be inflated by dominance and maternal effects rather than being only due to additive genetic effects (Falconer 1989).

We performed a set of analyses to test the effects of (1) beetle population, (2) genotype (family), (3) host plant (diet), (4) population by host interaction and (5) genotype by host interaction on growth rate, development rate, final size, and larval survival. Each performance trait was separately analysed with a mixed model analysis of variance (ANOVA), in which population, host and the population-host interaction were considered as being fixed effects, whereas family and the family-host interaction were considered random effects. Family was nested within population. We used type III sums of squares, which tolerate unbalanced sample sizes. Survival data were coded as zero (larva alive at third moult) or 1 (larval not surviving until third moult) as is standard practice in the quantitative genetics of threshold traits (Falconer 1989, Roff 1997).

Growth rates and development times were ln-transformed previous to analysis, to meet the ANOVA assumption of homogeneity of variances (Sokal and Rohlf 1995), whereas for the weight data this was not necessary. In line with the method described by Roff and Simons (1997) for quantitative genetics analysis of threshold traits we did not transformed the survival data.

Broad sense heritabilities of performance traits on each host in each population were calculated with the standard formula based on one-way ANOVAs performed separately for each host and population (Falconer 1989, Roff 1997). We tested whether heritabilities were significantly different from zero with t-tests based on standard errors calculated with the formula given by Roff (1997).

Genetic correlations between the two host species for each performance trait in each beetle population were estimated through Pearson product-moment correlations of family means (Via 1994, Roff 1997). This technique is widely used to calculate genetic correlations (Via 1994, Carrière and Roitberg 1995, Campbell 1997, Sgrò and Hoffman 1998) but is likely to overestimate them (Fry 1992, Roff 1997). In the same way, we estimated the genetic correlations between each pair of performance traits within each host for each population. Since the probability of finding a false significant correlation (type I error) increases with the number of tests that are simultaneously performed, we used sequential Bonferroni adjustments to calculate the significance threshold of each single correlation (Rice 1989).

Mean values of the performance traits are given $\pm$ their standard errors. Statistics were calculated with the JMP package (SAS 1989).

Results

Growth rate

The effect of host plant on the larval growth rate varied among beetle populations, as is stated by the significant statistical interaction between population and host (Table 1). Larvae from Mattmark had a higher mean growth rate when feeding on *C. spinosissimum* than when feeding on *A. alliariae*, whereas the opposite was true for the larvae from Lautaret (Fig. 1). In other words, Mattmark and Lautaret larvae grew faster on their native host genus than on the alternative novel host. Larvae from Petit St-Bernard had equal growth rates on both hosts (Fig. 1).

Overall, larvae from Mattmark grew faster than larvae from the other two populations, larvae from Petit

Table 1. ANOVAs for the performance traits. The family effect and the family-diet interaction were nested within population.

Trait Source of variation	df	MS	*F*	*P*
Growth rate:				
Population	2	6.5593	61.8957	<0.0001
Family (population)	72	0.1085	2.6811	<0.0001
Host	1	0.0327	0.8058	0.3721
Population × host	2	0.1460	3.5954	0.0321
Family × host (population)	72	0.0405	0.9289	0.6455
Error	1057	0.0436		
Development time:				
Population	2	12.0734	459.2192	<0.0001
Family (population)	72	0.0270	3.3751	<0.0001
Host	1	0.2902	36.2762	<0.0001
Population × host	2	0.0398	4.9784	0.0092
Family × host (population)	72	0.0080	1.0139	0.4481
Error	1063	0.0079		
Final size:				
Population	2	1248.49	38.3501	<0.0001
Family (population)	72	33.0186	2.0899	0.0010
Host	1	39.6531	2.4747	0.1196
Population × host	2	25.8892	1.6160	0.2051
Family × host (population)	72	15.7995	0.7398	0.9478
Error	1058	21.3562		
Survival:				
Population	2	0.2948	0.8957	0.4128
Family (population)	72	0.3295	2.1345	0.0008
Host	1	1.1157	7.2293	0.0089
Population × host	2	0.3233	2.0948	0.1305
Family × host (population)	72	0.1544	1.2642	0.0710
Error	1302	0.1221		

St-Bernard grew slowest and larvae from Lautaret had intermediate growth rates (significant population effect in Table 1; Fig. 1). Mean growth rates (± 1 SE) were 1.57 ± 0.021 mg/day for Mattmark larvae fed on *A. alliariae*, 1.62 ± 0.020 mg/day for Mattmark larvae on *C. spinosissimum*, 1.42 ± 0.025 mg/day for Lautaret larvae on *A. alliariae*, 1.35 ± 0.021 mg/day for Lautaret larvae fed on *C. spinosissimum*, 1.25 ± 0.016 mg/day for Petit St-Bernard larvae on *A. alliariae*, 1.23 ± 0.017 mg/day for Petit St-Bernard larvae fed on *C. spinosissimum*. The significant family (i.e. genotype) effect indicates the presence of genetic variation for growth rates (Table 1). The family-host interaction was not statistically significant.

Broad sense heritability of growth rate was statistically significant for Lautaret and Mattmark larvae growing on *A. alliariae* and for Petit St-Bernard larvae growing on *C. spinosissimum* and it varied roughly between 20 and 30% (Table 2). The non-significant heritabilities varied between 1 and 11% (Table 2).

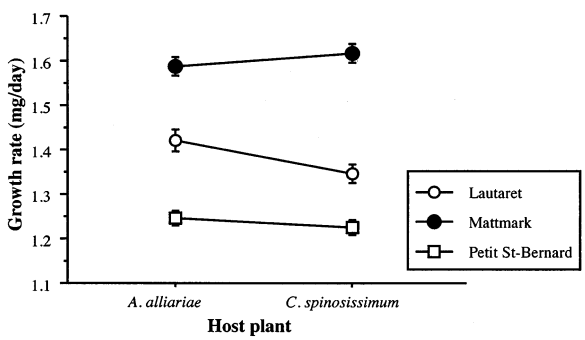

Fig. 1. Average larval growth rates (± 1 SE) of the three populations on both host plants.

Development time

Also in the case of development time, the effect of host depended on the beetle population (significant population-host interaction, Table 1). Larvae from both Lautaret and Petit St-Bernard had a longer mean development time when feeding on *C. spinosissimum* than when feeding on the other plant whereas larvae from Mattmark had about equal mean development times on both hosts (Fig. 2). Mattmark larvae needed 4–6 days less than larvae from the other two populations to reach the third moult (i.e. significant population effect in Table 1; Fig. 2). Larvae from Lautaret and Petit St-Bernard had about equal development times (Fig. 2). Again, there was a significant effect of family (Table 1) and mean development times were 14.1 ± 0.086 days for Mattmark larvae fed on *A. alliariae*, 14.2 ± 0.074 days for Mattmark larvae on *C. spinosissimum*, 18.8 ± 0.19 days for Lautaret larvae on *A. alliariae*, $19.5 \pm$

Table 2. Broad sense heritabilities of larval performance traits for each host and beetle population. *P*-values for differences from zero are given in parentheses.

Trait	Population		
	Lautaret	Mattmark	P. S. Bernard
Growth rate:			
A. alliariae	0.328 (<0.025)	0.191 (<0.05)	0.112 (>0.1)
C. spinosissimum	0.067 (>0.1)	0.012 (>0.1)	0.213 (<0.05)
Development time:			
A. alliariae	0.367 (<0.01)	0.188 (<0.05)	0.013 (>0.1)
C. spinosissimum	0.276 (<0.025)	0.349 (<0.01)	0.322 (<0.025)
Final weight:			
A. alliariae	0.162 (<0.1)	0.091 (>0.1)	0.066 (>0.1)
C. spinosissimum	−0.069 (>0.1)	−0.107 (>0.1)	0.004 (>0.1)
Survival:			
A. alliariae	0.808 (<0.005)	0.530 (<0.05)	0.695 (<0.025)
C. spinosissimum	0.474 (<0.025)	0.856 (<0.005)	0.940 (<0.001)

0.15 days for Lautaret larvae fed on *C. spinosissimum*, 19.0 ± 0.13 days for Petit St-Bernard larvae on *A. alliariae*, 19.7 ± 0.13 days for Petit St-Bernard larvae fed on *C. spinosissimum*.

Broad sense heritability of development time was statistically significant for all but one population-host combinations (Table 2). The statistically significant heritabilities ranged between 19 and 37% while the non-significant one, that for Petit St-Bernard larvae growing on *A. alliariae*, was slightly above 1% (Table 2).

Final weight

Final weights differed significantly among beetle populations (Table 1). One day after third moult, the larvae from Lautaret were the heaviest (mean 28.2 ± 0.28 mg) while the larvae from Mattmark were the lightest (mean 24.7 ± 0.21 mg) and larvae from Petit St-Bernard had intermediate weights (mean 25.6 ± 0.22 mg) (Fig. 3). The family effect was significant as well, whereas neither the host factor nor the interactions were statistically significant (Table 1).

Survival

Only the main factors family and host plant had a significant effect on larval survival; the interactions did not (Table 1). Larvae survived in higher proportion when feeding on *A. alliariae* than when feeding on *C. spinosissimum* (Fig. 4). Mean survival rates were 0.87 ± 0.013 for larvae raised on *A. alliariae* and 0.81 ± 0.015 for larvae raised on *C. spinosissimum*.

All *O. elongata* populations showed statistically significant broad sense heritability of survival on both host plants, with heritability values varying between 47 and 94% (Table 2).

Genetic correlations

Growth rate and development time showed significant, strong, positive correlations between hosts for Lautaret and Mattmark larvae but not for the Petit St-Bernard larvae, (Table 3). In other words, the genetic correlations across a host genus that is established and a host genus that is new to the beetle populations were significantly positive while the correlations between two es-

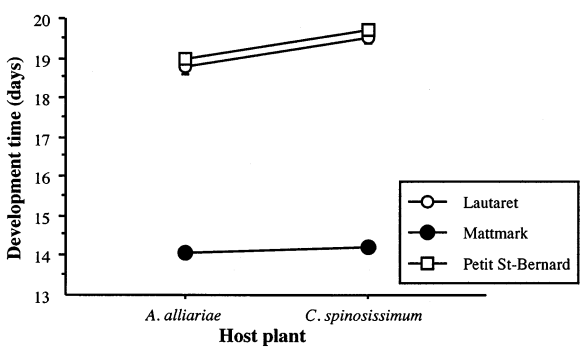

Fig. 2. Average development time (± 1 SE) to the third larval moult of three populations on both host plants.

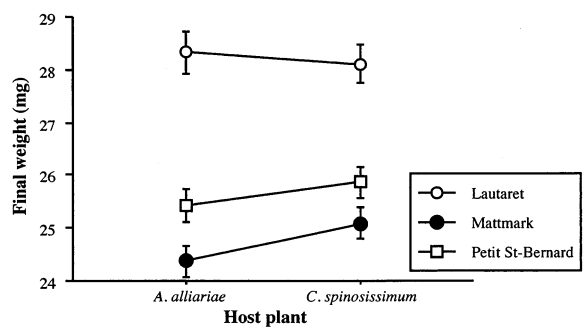

Fig. 3. Average weigh (± 1 SE) after the third larval moult of the three populations on both host plants.

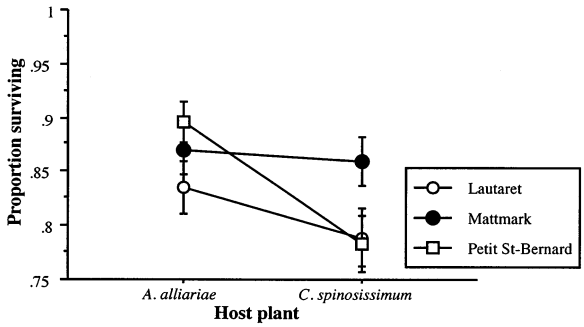

Fig. 4. Average survival to the third larval moult (± 1 SE) of three populations of on both host plants.

tablished hosts were not significantly different from zero. The across-host correlations for final weight and survival were not significantly different from zero (Table 3).

Discussion

Our results provide some support for the predictions of local adaptation in larval performance on the different hosts in these three populations of *O. elongata*. As predicted, the single host populations grew faster on their native host plant genus while the two-host population had equal growth rate on both host plants (significant population $\times$ host interaction, Fig. 1). However, the differences between host plant treatments within populations were small in all traits and the degree of local adaptation in these performance traits seems to be relatively limited. For the Lautaret population (native host *A. glabra*), the faster growth on *A. alliariae* did not influence the final weight but led to a slightly shorter larval period compared to *C. spinosissimum* (Fig. 2 and 3). For the Mattmark population the faster growth on the native host *C. spinosissimum* did not significantly influence either size at the third moult or the larval period compared to the *A. alliariae* case (Fig. 2 and 3). Larval growth rates of the population from Petit St-Bernard were the same for both plants but larvae reared on *A. alliariae* had a slightly shorter larval period compared to the siblings reared on *C. spinosissimum*. These patterns are partly in line with earlier studies of the Petit St-Bernard population although those studies have always indicated larger difference in larval growth rate between the plants with *A. alliariae* supporting the fastest growth (Ballabeni and Rahier 2000a, b).

In comparison to the relatively small effects of host plants the large differences between the populations in life history traits is the more striking (Fig. 1–3). For example, the Mattmark population grew faster on *C. spinosissimum* but it also had the highest growth rate of all populations on *A. alliariae* (Fig. 1). Indeed, the population from Mattmark describes a distinctly different life history compared to the other two populations by having a shorter larval period and typically ending up at a smaller weight (Fig. 2 and 3).

There was no general support for the predictions of lower heritabilities for performance traits on the native hosts compared to the alternative hosts (Table 2). Contrary to predictions, the estimates of broad sense heritabilities in the Lautaret population were in fact constantly highest on *A. alliariae*. At present we are unable to explain this but similar results was reported in laboratory study of the beetle *Callosobruchus maculatus* (Kawecki 1995). In the Mattmark population only the heritabilities of growth rate showed the expected lower value for the native host *C. spinosissimum* (Table 2).

The estimates of genetic correlations between the two host species were positive in all cases although none of them were significantly different from zero in the Petit St-Bernard population (Table 3). On the contrary, in the two single host populations the correlations for growth rate and development time were stronger and significantly different from zero. This pattern of stronger positive genetic correlations across host in the single-host populations as compared to the two-host population lends support to the equilibrium trade-off hypothesis put forward by Joshi and Thompson (1995). We did not find the predicted negative genetic correlation between hosts in the Petit St-Bernard population that is adapted to use both hosts. It is however important to note that this prediction rests on the assumption that the population has experienced selection in the two-host situation for a time period that allows a genetic equilibrium to be established. Indeed, the main feature of the equilibrium trade-off hypothesis is that

Table 3. Genetic correlations of each performance trait between the two hosts, estimated on family means by Pearson's product moment coefficients. *P*-values for differences from zero are given in parentheses. After sequential Bonferroni adjustments for multiple comparisons we accept only correlations in bold type as significant (table-wide alpha = 0.05).

Trait	Population		
	Lautaret	Mattmark	P. S. Bernard
Growth rate	**0.664** (0.0004)	**0.563** (0.0034)	0.095 (0.6432)
Developmental time	**0.671** (0.0003)	**0.709** (0.0001)	0.234 (0.2503)
Final size	0.471 (0.0200)	0.215 (0.3010)	0.238 (0.2420)
Survival	0.362 (0.0822)	0.374 (0.0655)	0.390 (0.0491)

negative genetic correlations between hosts evolve with time. At present, we have little information about the history of host plant availability in the three populations. It is possible that the combination of beetles with two hosts at Petit St-Bernard is relatively recent and that the beetle population has not reached a genetic equilibrium yet. The results on genetic correlations in our field-derived populations are similar to the results of a laboratory study of *Drosophila melanogaster* that was designed to test the equilibrium trade-off hypothesis (Joshi and Thompson 1997). The authors found that the genetic correlation across hosts (larval media) for development time was strongly positive in two populations that had been experiencing only one host for 12 generations, while in the replicated populations kept on both hosts during 12 generations the genetic correlation was zero rather than negative.

The present study agrees with the general finding in insect-host plant studies that larval performance trade-offs are unlikely to be the only or even the major evolutionary factor that limits the expansion of host plant range in herbivorous insects (se references in Joshi and Thompson 1995, 1997). Nevertheless, according to the equilibrium trade-off hypothesis it is possible that, with some additional time, the two-host situation at Petit St-Bernard will lead to the evolution larval performance trade-offs between the hosts. If so, we might see the evolution of specialization onto one of the hosts, or alternatively, the evolution of two host plant races at this site. However, when considering other fitness components in the system this scenario seems unlikely since the present knowledge suggests that at Petit St-Bernard natural selection is in fact favouring a dynamic utilization of both hosts during different parts of the life cycle (Ballabeni and Rahier 2000a, Ballabeni et al. 2001a, b). Moreover, the pattern in the single-host populations suggests that selection for specialization to one host leads to relatively minor improvement in larval performance on this host. Indeed, in *O. elongata* it seems as if most alleles that improve growth performance on one host also improves performance on the other host. A striking example of this is given by the Mattmark population that performs best on its native host *C. spinosissimum* but that also would outgrow the other populations on their native host genus *Adenostyles*. This would happen despite the fact that larvae in this population never encounter *Adenostyles* plants. We can only speculate about the reason for the peculiarities of the Mattmark population but it is possible that the seasonality is somehow different at this locality compared to the others, selecting for a faster development in general (Nylin and Gotthard 1998, Gotthard 2001). In this high alpine environment climatic variables may vary greatly within small distances since they are dependent on the orientation of slopes and peaks at the site as well as the closeness to glaciers, lakes and other water bodies. Another possibility is that since the native host of the Mattmark population, *C. spinosissimum*, does not provide any sequesterable PA:s the larvae may be more vulnerable to predation. Thus, compared to the other two populations there may be stronger selection favouring a high larval growth rate and a short larval period in the population at Mattmark. In any case, strong directional selection on faster larval growth in the Mattmark population is compatible with the very low heritability for growth rate on *C. spinosissimum* that were found in this population (Table 2). Such directional selection in combination with the strong positive genetic correlation for growth rate across host plants (Table 3) is a likely explanation for why the Mattmark population not only grows fast on its native plant but also on *A. alliariae*.

The spatial distribution of *O. elongata* and its host plants together with the low dispersal capacity of the beetles suggest that local adaptations in host-plant utilisation may evolve. It seems however that selection for local specialization have only led to a fairly limited divergence among populations in relative larval performance on the two hosts. Our study suggests that this can partly be explained by the genetic architecture of these traits. There are however strong indications that other aspects of host use in these populations have diverged more distinctly (Gotthard K., Magraf N., Rahier M. unpubl.). This may be due to different genetics of these traits or to a stronger correlation with individual fitness. In any case, we believe that continued focus on spatial variation and local adaptation in this relatively simple insect-host plant system will allow us to address additional questions that are relevant to the evolution of insect-plant interactions. Indeed, we fully agree with Thompson (1994) that an explicit geographical perspective will often be crucial for the understanding of the evolution of host plant specialization in insects.

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