



Evolution of plasticity prevents postinvasion extinction of a native forb

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Exotic plant invaders pose a serious threat to native plants. However, despite showing inferior competitive ability and decreased performance, native species often subsist in invaded communities. The decline of native populations is hypothesized to be halted and eventually reversed if adaptive evolutionary changes can keep up with the environmental stress induced by invaders, that is, when population extinction is prevented by evolutionary rescue (ER). Nevertheless, evidence for the role of ER in postinvasion persistence of native flora remains scarce. Here, I explored the population density of a native forb, *Veronica chamaedrys*, and evaluated the changes in the shade-responsive traits of its populations distributed along the invasion chronosequence of an exotic transformer, *Heracleum mantegazzianum*, which was replicated in five areas. I found a U-shaped population trajectory that paralleled the evolution of plasticity to shade. Whereas *V. chamaedrys* genotypes from intact, more open sites exhibited a shade-tolerance strategy (pronounced leaf area/mass ratio), reduced light availability at the invaded sites selected for a shade-avoidance strategy (greater internode elongation). Field experiments subsequently confirmed that the shifts in shade-response strategies were adaptive and secured postinvasion population persistence, as indicated by further modeling. Alternative ecological mechanisms (habitat improvement or arrival of immigrants) were less likely explanations than ER for the observed population rebound, although the contribution of maternal effects cannot be dismissed. These results suggest that *V. chamaedrys* survived because of adaptive evolutionary changes operating on the same timescale as the invasion-induced stress, but the generality of ER for postinvasion persistence of native plants remains unknown.

biological invasions | evolutionary rescue | phenotypic plasticity | shade-response strategies

Introductions of exotic species or shifts in species ranges due to global warming generate novel species assemblages (1–4). Since these introduced biota lack a coevolutionary history with the invaded community, they can negatively affect resident plant and animal populations, particularly after the introduction or expansion of pests, parasites, and predators (2, 5). Novel competitors can have similarly detrimental effects on resident plants (4, 6), although examples of large-scale extinctions due to invasive or expansive plants are quite rare (7–10).

Some researchers have argued that the time since introduction is still too short to observe the full impact of plant invasions on resident plants, that is, they assume plant invaders induce an extinction debt in the native flora (11). Such assumptions can, however, be too simplistic, given that ecological and evolutionary processes can modify the long-term effects of invasive plants on native flora. Indeed, while these effects can intensify over time to the larger detriment of native flora, they can also, in contrast, lessen over the invasion history, thereby favoring the recovery and persistence of native populations (12–15). As an example of the latter scenario, the decline of native populations can be halted and eventually reversed if adaptive evolutionary changes can keep up with the environmental stress induced by invaders, namely when extinction is prevented by an evolutionary rescue (16–19). Nevertheless, proof of evolutionary rescue from the wild remains scarce and elusive, largely because the appropriate combination of demographic and adaptive phenotypic evidence is usually not available (17). Moreover, population recovery can involve a number of alternative mechanisms, including habitat improvement, a plasticity-mediated increase in individual fitness, or the addition of immigrants (19–21), which make validation of evolutionary rescue in natural conditions even more difficult.

The giant hogweed *Heracleum mantegazzianum* is a problematic exotic weed worldwide (22) that reduces native grassland diversity (23) by forming extensive and dense stands (Fig. 1A) with stems up to 5 m tall, thus largely reducing light availability (24). However, its influence on native diversity can diminish over time as its cover declines and light availability increases in the more advanced phases of invasion, likely because of the accumulation of specialized soil pathogens (15). In this study, I used an invasion chronosequence of

Significance

The potential of evolutionary rescue (ER) to prevent population extinctions in deteriorating environments is increasingly being appreciated. However, evidence suggesting that ER underlies the postinvasion persistence of native flora remains scarce. By exploring a native forb's population density and trait changes along a replicated invasion chronosequence, I found a U-shaped population trajectory that paralleled the evolution of plasticity to shade. Whereas genotypes from intact, more open sites exhibited a shade-tolerance strategy, the reduced light availability at invaded sites selected for a shade-avoidance strategy. These shifts in shade-response strategies were adaptive and secured postinvasion population persistence. The findings suggest that native populations may survive because of adaptive evolutionary changes operating on the same timescale as the invasion-induced stress.

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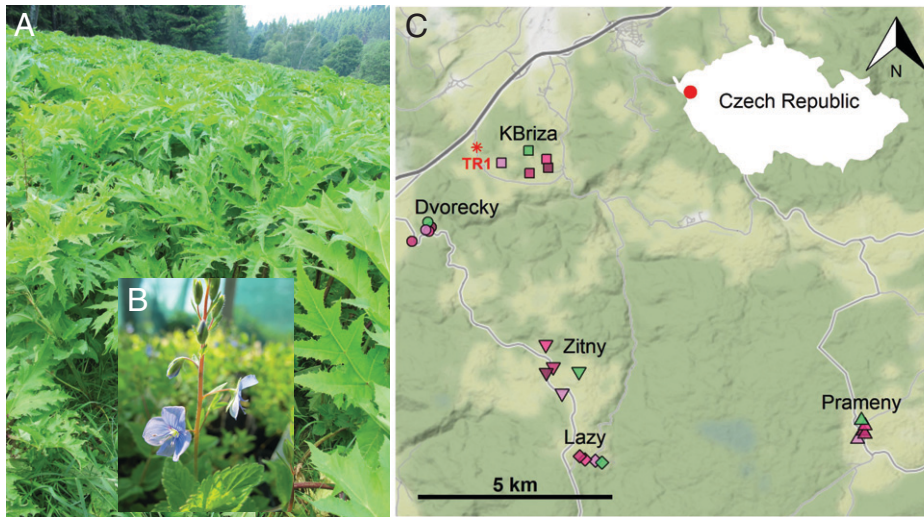


Fig. 1. Exotic umbellifer *H. mantegazzianum* (A), native forb *V. chamaedrys* (B), and sites with different invasion histories included in this study (C). In each of the five areas (located in the west of the Czech Republic; C, Inset), an unin invaded site (in green) and sites invaded for ~11, 28, 42, or 48 y (from light to dark violet) were surveyed for the population density of *V. chamaedrys* and served as a source of the forb's genotypes used in the garden and field experiments (the latter were conducted at the TR1 site).

giant hogweed (including an unin invaded site and sites invaded for ~11, 28, 42, or 48 y) that was replicated in five areas (Fig. 1C and *SI Appendix*, Fig. S1) to explore the population density of the native grassland forb *Veronica chamaedrys* (Fig. 1B) at different invasion stages. I then evaluated whether the observed population recovery of the forb at sites where the invader had been present for longer durations could be attributed to evolutionary rescue or to alternative (ecological) mechanisms. Specifically, I investigated whether the rebounding population density of *V. chamaedrys* in advanced invasion phases could be explained by improved habitat conditions, such as elevated light or soil nutrient availability. I also tested whether the local density could be associated with *V. chamaedrys* abundance at the landscape level, which would suggest that immigration [i.e., demographic rescue (19)] was responsible for the observed recovery. While attempting to determine whether evolutionary rescue was a more likely mechanism of the recovery, I conducted a series of common garden and field experiments complemented by modeling. In the common garden with vegetatively propagated plant material of *V. chamaedrys* collected across different invasion phases, I searched for phenotypic population differentiation in two shade-responsive traits. The same plant material was then used in the field experiment to test whether the differentiation is adaptive. Finally, using the data for the phenotypic differentiation obtained in the common garden experiments and of the vital rates derived from the field experiment, I modeled the population growth of *V. chamaedrys* across environmental (light) conditions typical for distinct invasion phases. I was particularly interested in determining whether the evolutionary trait differentiation is likely to restore positive population growth under the light conditions most altered by the invasion. In addition, I decomposed the contributions of ecological (light availability) and evolutionary (trait evolution) changes to variations in population density recorded across different invasion stages (an overview of the experiments and synthesis is provided in *SI Appendix*, Fig. S2).

Results

Ramet density of the native forb reduced greatly in the early invasion stages (from 5.91 ± 2.46 ramets per 625-cm^2 area [mean \pm CI] in unin invaded sites to 1.76 ± 1.65 ramets per 625-cm^2 area in sites invaded for ~11 y), but it started to

rebound later, reaching a density of 4.51 ± 3.57 ramets after 48 y of invasion (Fig. 2A; time: $t = -2.691$, $P = 0.010$; time²: $t = 2.831$, $P = 0.008$; *SI Appendix*, Table S1). This recovery cannot be explained by immigration, since the local ramet density was unrelated to the regional abundance of the species (*SI Appendix*, Materials and Methods, Fig. S3, and Table S2), nor can it be accounted for by improved environmental conditions in the later invasion stages. The light availability (defined as the proportion of photosynthetically active radiation [PAR] reaching the ground level, Δ PAR) that substantially reduced at sites invaded for 11 y indeed increased with advancing invasion time, although it never reached preinvasion values (Fig. 2B; time: $t = -3.108$, $P = 0.009$; time²: $t = 2.865$, $P = 0.015$; *SI Appendix*, Table S1; see also ref. 24). Ramet density at invaded sites was, however, unrelated to Δ PAR (Fig. 2C; $t = -0.017$, $P = 0.987$; *SI Appendix*, Table S1) or to the measured soil characteristics (*SI Appendix*, Table S1).

Therefore, I searched for a postinvasion adaptive evolutionary response that would support evolutionary rescue as a more plausible mechanism of recovery. As a part of this investigation, I set up a common garden experiment with five genotypes from each study site (i.e., 115 genotypes from 23 populations in total; *SI Appendix*, Table S3) grown under two shading levels, which corresponded to light conditions at intact and invaded sites, respectively (set up with 30 and 70% shade cloths; *SI Appendix*, Fig. S4). I used this setup since I expected population differentiation in phenotypic plasticity to depend on the extent of the reduction in light availability at the source sites (25–27). Specifically, plants can tolerate a reduction in PAR levels by increasing light-capture efficiency, for example, by increasing leaf area at the expense of leaf thickness (thereby increasing the specific leaf area [SLA]) (25). This strategy may, however, be maladaptive at very low light levels since the large leaves produced at little construction costs are more sensitive to mechanical stress and herbivory (28). Plants from the invaded populations, that is, from sites with very low PAR levels, were therefore expected to display a shade-avoidance strategy instead. This strategy allows plants to grow away from inferior light conditions and forage for better conditions through leaf and stem elongation, including horizontal spread via increased internode length (IL) (27).

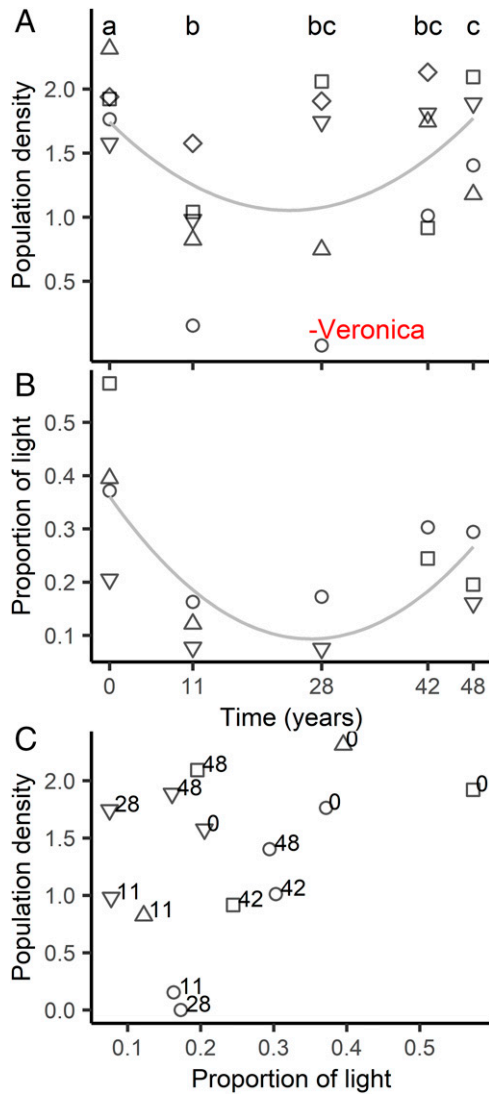


Fig. 2. Population density and light conditions along the invasion chronosequence. (A) Ramet density of *V. chamaedrys* (log number of ramets per 625-cm² area) from five areas (symbols refer to the five areas as in Fig. 1) plotted against the invasion history of *H. mantegazzianum*. Significant differences between invasion times are indicated by different letters; the gray line shows a quadratic fit when time was a continuous predictor. Although the species persisted at most sites, it was absent from one locality (-Veronica), possibly going extinct due to invasion. (B) The proportion of PAR reaching the ground level in different invasion phases with a quadratic fit (time = 0 y indicates uninvasion sites). (C) Scatterplot of ramet density (with invasion time) against proportion of light.

The plasticity of IL in response to stronger shading was indeed larger in populations from invaded than from uninvasion sites (0.313 ± 0.022 vs. 0.260 ± 0.029 ; $t = 2.632$, $P = 0.010$). Pronounced internode elongation was not, however, favored over the whole course of invasion. An increase in IL plasticity in populations in the initial invasion phases (until ~ 28 y of invasion) was reversed in the more advanced stages of invasion (Fig. 3A; time: $t = 2.767$, $P = 0.006$; time²: $t = -2.549$, $P = 0.011$; SI Appendix, Table S4). Consistent with my initial expectation, I also found that plasticity in the SLA was smaller in the invaded than in the uninvasion populations (0.335 ± 0.015 vs. 0.364 ± 0.026 ; $t = -2.191$, $P = 0.033$). I also found that SLA plasticity changed, albeit nonsignificantly ($P \leq 0.05$), during invasion history, in the opposite direction of IL plasticity (Fig. 3B and SI Appendix, Table S4). The variations in the plasticity of the two traits can be explained by the light conditions at the genotypes'

source sites. While a larger Δ PAR (which is characteristic for uninvasion sites and sites at more advanced invasion stages; Fig. 2B) favors larger SLA plasticity ($t = 2.996$, $P = 0.012$), it has an opposite effect on IL plasticity ($t = -2.798$, $P = 0.017$; Fig. 3C). The variations in the mean values and the plasticity of the two traits (and of fitness traits) are heritable, although the heritability varied nonsignificantly across the three garden experiments (SI Appendix, Table S5). Moreover, the contribution of transgenerational (nongenetic) effects of maternal environment on the observed differences in the two traits cannot be ruled out (29). Although the mean IL values of the same genotypes persisting for increasing time periods (for 2, 5, and 6 y prior to the experiment) in uniform light conditions were similar, the mean SLA values as well as trait plasticity varied to a larger extent across the

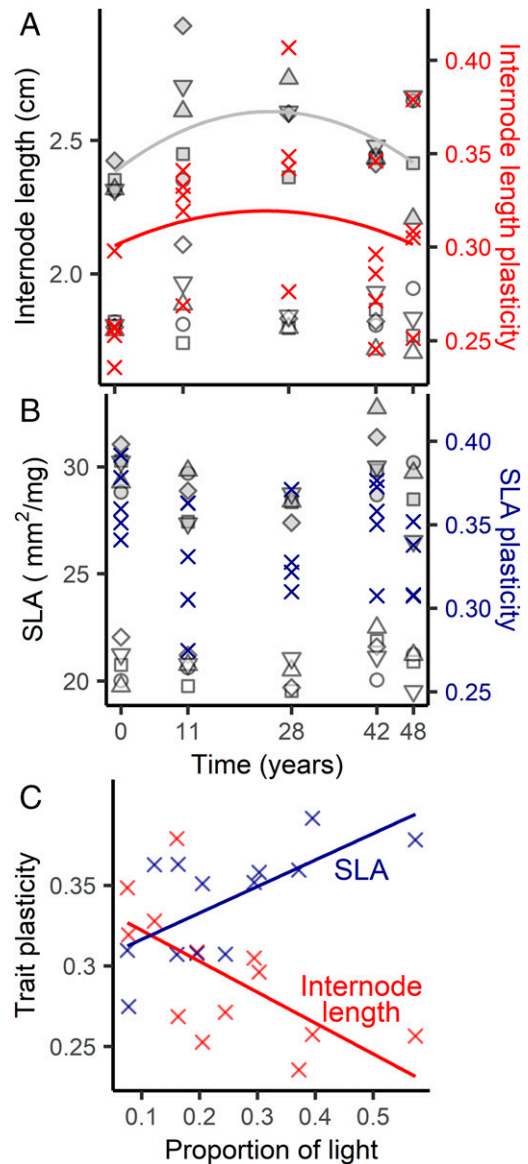


Fig. 3. Evolution of shade-induced trait plasticity. (A and B) Internode length (A) and specific leaf area (B) of populations (based on five genotype-specific means per population) from sites differing in invasion history (symbols refer to the five areas in Fig. 1). The open and full symbols refer to cultivation under 30 and 70% shade cloth, respectively. Trait plasticity is depicted by crosses and was calculated as the log response ratio of trait means measured for plants under the two shading levels. A significant effect of invasion history is indicated by a quadratic fit. (C) Significant associations between trait plasticity measured in the garden experiment and the proportion of light reaching the ground levels at the genotypes' source sites.

three garden experiments (*SI Appendix, Table S6*). I also found that the plasticity in the two traits could evolve independently in each trait, given the nonsignificant genetic correlation between the two (*SI Appendix, Table S7*).

In a field experiment, I then examined the adaptive value of the garden-observed variation in plasticity in the two shade-responsive traits. The same genotypes as used in the garden experiment (*SI Appendix, Table S3*) were planted at an invaded site, in plots treated by clipping and plots left intact (Fig. 1C). Plants originating from uninvaded sites survived better in the clipped plots, while the other plots promoted survival of plants from invaded sites (invasion experience \times clipping treatment: $z = 2.350$; $P = 0.019$; *SI Appendix, Fig. S5 and Table S8*). I also confirmed that the elevated SLA plasticity was adaptive at high Δ PAR values, as indicated by a significantly positive effect of their interaction on plant survival (SLA plasticity \times Δ PAR: $z = 2.482$, $P = 0.013$; Fig. 4A and *SI Appendix, Table S9*). In contrast, the high IL was adaptive at low Δ PAR values, as indicated by a significantly negative effect of the interaction on plant size (IL plasticity \times Δ PAR: $t = -1.985$, $P = 0.046$; Fig. 4B and *SI Appendix, Table S9*). Although the findings proved that changes in SLA and IL plasticity along a light availability (Δ PAR) gradient were adaptive, they did not clarify whether the observed evolution of trait plasticity could restore positive population growth and thus prevent the extinction of *V. chamaedrys* at very low Δ PAR values, namely in light conditions typical for the early invasion stages. To test this aspect, I investigated the population growth rates (λ s) in different light conditions by

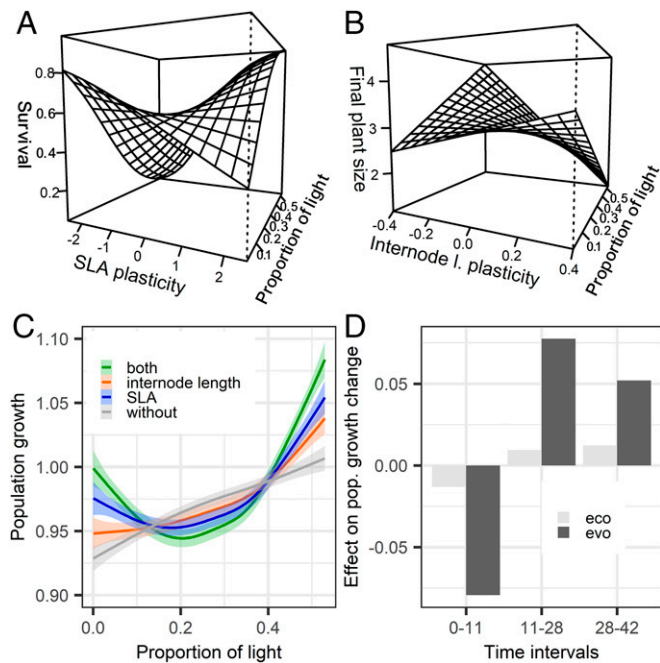


Fig. 4. Adaptive value of trait plasticity (estimated in the 2011 to 2013 garden experiment) measured in the field (2012 to 2013). (A) Survival probability as a function of SLA plasticity (BLUP values) and the proportion of light reaching the ground level in the respective experimental plots. (B) Final plant size (log mg) as a function of plasticity in internode length (BLUP values) and the proportion of light in the plots. (C) Results of integral projection models showing the effect of the proportion of light on the population growth rate. No evolution (without) and evolution of shade-induced plasticity (in SLA and IL only, or in both traits according to the association in Fig. 3C) were included to assess the effect of light availability on λ . Curves were drawn using generalized additive models (mean \pm 95% CIs). A Δ PAR of 0.4 corresponds to average light conditions at uninvaded sites. (D) Contribution of changes in light availability (eco) and of evolution of IL plasticity (evo) to the field-observed change in population growth [$\log(\lambda)$].

using integral projection models (IPMs) with or without evolution of shade-induced plasticity (*SI Appendix, Fig. S6*). In the IPMs, I integrated all vital rates while considering the effects of trait plasticity \times Δ PAR interactions on survival and growth (*SI Appendix, Tables S9 and S10*). I found that *V. chamaedrys* populations could indeed survive (with $\lambda \geq 1$) at very low Δ PAR values, but only if the plasticity in both traits evolved along the Δ PAR gradient (Fig. 4C; note that λ was ≥ 1 for a narrow range of Δ PAR values around 0.05 and smaller; simulated λ values are shown in *SI Appendix, Fig. S7*). This finding, together with the large relative contribution of plasticity evolution to the field-observed variation in population growth (Fig. 4D and *SI Appendix, Fig. S8*), suggests that evolutionary rather than ecological processes determine the observed population trajectory and postinvasion survival of *V. chamaedrys*.

Discussion

This study documents a mechanism that was hypothesized but has been scarcely recorded in the wild, namely the ability of rapid evolutionary changes to rescue resident populations from extinction under anthropogenic stress. I showed that the novel competitor *H. mantegazzianum* causes a much stronger resource shortage than residents, which manifested through a significant decrease in light availability. Nevertheless, even though these effects drive *V. chamaedrys* populations close to extinction, they eventually persist thanks to evolutionary shifts from a shade-tolerance (pronounced leaf area/biomass ratio, SLA) to a shade-avoidance strategy (greater internode elongation).

Native species have been documented to be displaced by invasive competitors (23), but they are also often reported to co-occur with the invaders (30, 31), despite their inferior competitive ability and decreased performance in the invaded communities (31, 32). This phenomenon has been attributed to the large initial population sizes, long life span of perennials (or the persistent soil seed bank of some annuals), as well as the capacity of some life stages to withstand the novel environmental conditions, all of which generate temporal lags in the extinctions of native plants (11, 33); thus, extinctions can be fully observed only long after invasion. However, most long-term predictions of native populations' prospects do not consider evolutionary responses to invaders and the resultant possible changes in vital rates affecting the population dynamics, possibly yielding misleading forecasts. Indeed, modeling the λ of *V. chamaedrys* based on the vital rates of uninvaded phenotypes predicted that its populations would perish under invasion-transformed conditions, namely under very low light availability. However, field observations showed the forb persisted and rebounded from decline, with the rebound occurring despite the harshest light conditions around the 30th year of the invasion (Fig. 2 A and B). However, the model provided more realistic estimates of population growth under a range of light conditions only when the demographic performance of phenotypes differentiated along an invasion-driven Δ PAR gradient was considered. These findings illustrate that potential evolutionary changes should not be neglected when predicting the impact of invaders on native population persistence.

The evolution of shade-induced plasticity supported the persistence and recovery of the native forb in the invaded communities. Plants have been documented to adjust their plastic responses depending on the height and density of competing neighbors; thus, tall and dense neighbors may elicit distinct morphological responses in comparison with short and sparse competitors (26, 27). Although the adaptive value of different plastic responses

under distinct competitive (shading) environments has been usually assumed in these previous experiments, it has been rarely tested. The findings in this study proved that greater internode elongation, but a less pronounced SLA increase, is adaptive under very low PAR, namely conditions typical for invaded sites (Fig. 4 *A* and *B*). This also explains an increasing trend in IL plasticity with a decreasing SLA plasticity of genotypes from sites with strongly reduced light availability (Fig. 3 *C*), which indicated minimal inter-annual variations in light conditions and consequently in selection regimes at sites with a distinct invasion history. However, as the light conditions changed nonlinearly over the course of invasion (Fig. 2*B*), most likely because the invader declined in the later invasion phases (15), so did the direction of selection, favoring plastic responses typical for uninvaded sites (Fig. 3 *A* and *B*). Thus, the boom-and-bust cycles of invader populations can be also assumed to have long-term consequences for the genetic diversity and the adaptive capacity of evolutionarily responsive native populations (e.g., ref. 34), deserving attention in future research.

Despite my assumption that the increase in the frequency of genotypes with adaptive plastic responses would underlie the observed rebound in the population densities, I could not entirely discount alternative explanations of the demographic trajectory. To evaluate these possible alternative explanations (19–21), I investigated the association of the density with improvements in light (and soil) conditions in the later invasion phases and with the species abundance in the surroundings; however, these investigations did not reveal any significant relationship (*SI Appendix*, Tables S1 and S2). Notably, I used space-for-time substitution to explore the population dynamics of the forb, light availability, and evolution of shade-induced plasticity; thus, I cannot eliminate the possibility of existing preinvasion differences in these parameters. This is, however, very unlikely since the data were collected in independent invasion chronosequences replicated in five areas (Fig. 1 *C*). Further, adaptive genetic changes constitute the basis of evolutionary rescue (17), but I cannot dismiss the possibility that nongenetic effects were also involved. I tried to minimize these effects by keeping plant material for 2 y in uniform light conditions prior to the main experiments, which was a substantially longer period than is typically used (35). Stability in genotypic means in IL values after up to 6 y in uniform conditions further suggested limited effects of maternal environment (*SI Appendix*, Table S6). Support for the involvement of nongenetic effects was noted, however. Specifically, the contribution of maternal environmental effects to the observed plasticity differentiation was suggested by the minimal congruence in trait plasticity of the same genotypes across the three experiments (*SI Appendix*, Table S6) as well as evidence showing that the predicted evolutionary response in trait plasticity was always smaller than the observed response (*SI Appendix*, Fig. S9). Thus, differences in intragenerational plasticity of genotypes from distinct invasion phases likely were due to both genetic and additional epigenetic differentiation, yet the relative contribution of each of the two mechanisms can be estimated only by using genetic and epigenetic markers (36). Another limitation of the study is that the adaptive value of plasticity differentiation and the vital rates used in modeling were derived from a short-term field experiment conducted at a single site. Finally, despite the evidence presented in this study, evolutionary rescue may not be a general mechanism for maintaining the postinvasion persistence of native populations, since, in addition to *V. chamaedrys*, very few plant species survived giant hogweed's invasion in the study system, specifically in the invasion phases with the harshest light conditions (15).

Materials and Methods

A detailed description of the materials and methods can be found in *SI Appendix*.

Field Surveys. To study the effect of invasion and its temporal dynamics on *V. chamaedrys*, the population density of *V. chamaedrys* was measured at 24 sites with differing invasion histories of *H. mantegazzianum* in the Slavkovský les Nature Reserve (Fig. 1*C* and *SI Appendix*, Fig. S1). These 24 grassland sites are described in Dostál et al. (15) and are distributed among five areas (Dvorecky, Kostelni Briza, Lazy, Prameny, Zitny). Each area included an uninvaded site and sites invaded for ~11, 28, 42, or 48 y, yielding 24 sites in total (since a site invaded for 48 y was not identified in the Lazy area). The population density of *V. chamaedrys* was measured at all 24 study sites in June 2011 during a vegetation survey described in ref. 15. At each site, I counted and measured the length of all *V. chamaedrys* ramets present in 18 (or 12) plots of 0.25 × 0.25 m. These plots were placed within 9 (or 6) larger plots of 2 × 2 m (i.e., two 0.25 × 0.25 m plots per large plot), which were set up to record species composition and placed along three (or two) transects 7 m apart (15).

To evaluate the light availability changes along the invasion chronosequence, PAR was measured in 20 out of the above-described 24 sites (the Lazy area was omitted) in August 2012 (results were obtained from ref. 24). At each study site, two transects per site (14 m apart) with three 2 × 2 m plots per transect were set up (not identical to the transects used for *V. chamaedrys* sampling but located at a distance of <100 m). In each plot, five measurements of PAR availability were taken in five randomly selected positions 5 cm aboveground and five measurements were obtained in the same positions at 20 cm above vegetation. The proportion of transmitted light was then calculated as a ratio of PAR amounts (Δ PAR) at the two heights, and was averaged across the five positions within plots and then across plots in a site. Light measurements from six study sites had to be excluded from further analyses since eradication of the invader started in 2011 in the study area (24). In addition to the light measurements, several soil characteristics were analyzed: Conductivity, pH, total nitrogen and carbon contents (and their ratio), and the amount of extractable phosphorus were measured in one composite soil sample per site after pooling six or nine 100-cm³ soil samples collected in species composition plots (15). The soil samples were collected in 2011, and the detailed findings of soil analyses are provided in the study by Jandová et al. (24). To estimate the abundance of *V. chamaedrys* at the landscape level, a vegetation survey was conducted in late May 2013 at eight 5 × 5 m plots located along cardinal directions at distances of 100 and 500 m from each study site (*SI Appendix*, Fig. S3). The percentage cover was recorded for each species present in the plots, and *V. chamaedrys* presence was then extracted from these records for further analyses.

Common Garden Experiments. To explore whether *V. chamaedrys* populations from sites with different histories differ in their shade-response strategies, I conducted a series of common garden experiments with *V. chamaedrys* genotypes exposed to different shading levels. For these experiments, 20 rooted ramets of *V. chamaedrys* were sampled at 23 of 24 study sites (the species was missing from one site in the Dvorecky area that had been invaded for 28 y) in early June 2010. The ramets were sampled at a distance of at least 5 m from each other to ensure that each ramet represented a different genotype (confirmed by isoenzyme genotyping described in *SI Appendix*, *Materials and Methods*). Ramets were grown in the experimental garden under 30% shade cloth until July 2011. For the shading experiment, I randomly selected five genotypes per population that were further propagated by stem cuttings and grown under 30% shade cloth until July 2012. Please note that root stimulant (based on hormones from the auxin class) was used during propagation, which could have side effects on plant phenotypes. However, rooted stem cuttings were employed in the experiment 1 y after the stimulant was applied, that is, these side effects likely had dissipated.

The experiment consisted of growing plants under two different shading levels (with eight plants per level of each genotype), which was achieved using 30 and 70% green shade cloths installed at a height of 1 m above the plants (July to October 2012 and April to July 2013; *SI Appendix*, Fig. S4). Additional light measurements indicated that the two shading treatments corresponded well to the Δ PAR recorded at uninvaded and invaded sites, respectively (*SI Appendix*, *Materials and Methods*). From the initial 1,809 plants (23 populations × 5 genotypes × 16 plants = 1,840, but fewer cuttings were available for

10 genotypes), 1,794 plants survived until June 2013. In June 2013, I sampled one leaf from each experimental plant and measured its area (LI-3100 area meter; LI-COR). The leaves were then dried to a constant weight and weighed, allowing determination of the SLA (mm^2 leaf area/mg leaf mass). In mid-July 2013, I terminated the experiment by harvesting the aboveground biomass and separated the vegetative and generative structures of each plant. Prior to the harvest, the number of ramets as well as the length and number of internodes of the longest ramet were measured for each plant; the latter two parameters were then used to calculate the mean IL (in cm). In the analysis of evolution of shade-responsive traits, I used the SLA and mean IL (25).

Two additional shading experiments were conducted during 2014 to 2016 and 2015 to 2017 by using the same plant material, but with increasing time periods in uniform conditions (kept under 30% shade cloth). These experiments were conducted to determine whether the population differentiation in shade-responsive traits observed in the first shading experiment (2011 to 2013) was determined genetically or by maternal light environment at the respective source sites (29).

Field Experiment. I performed a field experiment to test whether the population differentiation in the plastic responses of SLA and IL to experimental shading (Fig. 3C) represented adaptive strategies to distinct environmental (light) conditions. In May 2012, I multiplied the same genotypes as those included in the garden experiments. In early November 2012, the rooted cuttings were planted at two invaded sites (see the TR1 and TR2 sites in *SI Appendix, Fig. S1*) but outside the genotype source sites. At each site, the cuttings were planted in two plot types, which were treated by biomass removal (by clipping) and by leaving biomass intact, respectively. Treatment was performed prior to planting (with nine and eight plots per treatment type at the TR1 and TR2 sites, respectively). Each genotype (out of the 75 genotypes; *SI Appendix, Table S3*) was represented by at least two rooted cuttings per plot type at both experimental sites. At the TR1 and TR2 sites, I initially planted 340 and 370 cuttings, respectively, and recorded their initial sizes (number of ramets and length of the longest ramet).

Survival of the plants was evaluated in April 2013. In late May 2013, the plots assigned for clipping at the TR1 site were clipped, including the planted *V. chamaedrys*. Due to a miscommunication with the landowner, the TR2 site was mown completely in early May 2013. Although the plants were checked and measured in a subsequent census, the data from the TR2 site were not used in further analyses due to this disturbance.

In late June 2013, plant survival was recorded again. In the surviving plants, the following parameters were also measured: number of ramets, length of the longest ramet, and number of internodes of the longest ramet. I also harvested the biomass of surviving plants (subsequently dried to a constant weight) and counted the number of seeds for each fruiting plant. During the harvest, a total of 178 plants of 66 genotypes were obtained at the TR1 site (*SI Appendix, Table S3*). Prior to the harvest (in June 2013), I measured the light conditions for each plot in five positions, similar to the PAR measurements described above. At the TR1 site, clipped plots were characterized by a mean $\Delta\text{PAR} = 0.205 \pm 0.067$, which was significantly larger than the $\Delta\text{PAR} = 0.046 \pm 0.033$ in nonclipped plots ($n = 18$ plots, $t = -4.938$, $P \leq 0.001$).

Statistical Analyses. All mixed-effect models were developed using the lme4 (37) package of R (38), if not specified otherwise.

Effect of invasion history on *V. chamaedrys* population density and light availability. I tested for the effect of different invasion histories of *H. mantegazzianum* on the population density of *V. chamaedrys* [ratio of ramet number divided by sampled area; $\log(x + 1)$ -transformed]. For this analysis, the age of the hogweed populations (i.e., uninvaded sites and sites invaded for 11, 28, 42, or 48 y) was entered as a fixed effect (as a categorical predictor) and the study area (i.e., Dvorecky, Kostelni Briza, Lazy, Prameny, or Zitny) was entered as a random factor. I then used the multcomp package of R (39) to test for significant differences between time categories.

I also used an alternative model in which invasion time served as a continuous predictor. Specifically, I searched for a U-shaped curve of population dynamics that is typical for evolutionary rescue (17, 18) by testing for both linear and quadratic terms for invasion time (entered as fixed effects). Area was included as a random factor. Uninvaded sites were assigned residence time = 0 y. The significance of fixed effects was tested by a likelihood-ratio test.

I then tested for the effect of invasion history on ΔPAR at the study sites. In this analysis, I used simple linear regression instead of a mixed-effect model due to the smaller number of measurements ($n = 14$) obtained from the four areas. I included both linear and quadratic terms for time to test whether the decreased light availability that is a characteristic of the initial invasion stages can rebound in more advanced stages of invasion (see also refs. 15 and 24).

Finally, I used simple linear regression with population density as the response variable and ΔPAR as the predictor. I developed two models, one each with and without uninvaded sites. I also analyzed the relationships between population density and soil characteristics. All results are summarized in *SI Appendix, Table S1*.

Effect of landscape-scale abundance on the population density of *V. chamaedrys*. I tested whether variations in the local population density of *V. chamaedrys* are associated with the species abundance at the landscape level. For this analysis, I linked the population density at the respective study site with the presence of the species in surveyed plots within the area while taking into account the distance to these plots. Typically, each area contained five study sites (uninvaded and invaded for 11, 28, 42, or 48 y), and each of them was surrounded by eight plots (*SI Appendix, Fig. S3*) that were surveyed in 2013 (45 plots or sites per area in total). The population density of *V. chamaedrys* in a site was thus related to its presence or absence in 44 plots per area by calculating the connectivity index (40). I then developed mixed-effect models with the local population density of *V. chamaedrys* as the dependent variable and the connectivity index as the fixed effect. Area was used as a random effect (*SI Appendix, Table S2*).

Effects of experimental shading and invasion history on shade-responsive traits of *V. chamaedrys*. I tested for the effect of invasion and invasion history on the shade-responsive traits and their plasticity. For these analyses, I calculated the genotypic mean of SLA and IL separately for experiments conducted with the 30 and 70% shade cloths (based on eight replicates per genotype for each shading level), respectively. These mean values were then used to calculate genotype-specific trait plasticity as a log response ratio, $\log RR = \log \frac{\bar{X}_{70\%}}{\bar{X}_{30\%}}$ (41). From these genotypic mean (or log RR) values, I then calculated the population means, based on five genotypes per population.

First, I searched for the effect of invasion (presence of the invader at a site) on the plasticity in SLA and IL. Next, the population means in SLA and IL and in-trait plasticity were related to the invasion history. For this analysis, I used linear mixed-effect models with time entered as a fixed effect and area as a random effect. I used two sets of models, the first with time entered as a linear term alone, and the second with both linear and quadratic terms for time. The statistical significance of the fixed effects was estimated by likelihood-ratio tests, and the best-fitting model was identified on the basis of Akaike's information criterion. Finally, I searched for the association between the population-specific plasticity of the two traits and the ΔPAR at the corresponding source sites by using simple linear regression (*SI Appendix, Table S4*).

Heritability of shade-responsive traits and the fitness of *V. chamaedrys*. I analyzed heritable variations in five phenotypic traits, including three fitness characteristics: SLA, IL, total biomass, number of ramets, and number of seeds. I used the method described by Zhang et al. (42) to obtain the broad-sense heritability for each trait and its plasticity. I also performed separate calculations for the three common garden experiments (*SI Appendix, Table S5*).

Next, I searched for correlations between the genotype-specific mean values and the plasticity in SLA measured across the three experiments, but under the same shading level or with combinations of both shading levels. I determined similar correlations for IL measurements obtained in the three experiments (*SI Appendix, Table S6*). Finally, I evaluated the genetic correlations between SLA and IL (of trait means and plasticity) within the individual common garden experiments (*SI Appendix, Table S7*). In both correlation analyses, I used the best linear unbiased predictor (BLUP) approach (43, 44).

Adaptive value of plasticity in the shade-responsive traits tested in the field experiment. In all analyses, I used data from the TR1 site alone due to the previously described disturbance at the TR2 site. First, I searched for differences in survival (as of June 2013) between plants with and without previous invasion experience in the clipped and nonclipped plots. I used a generalized mixed-effect model with clipping treatment, invasion experience, and their interaction as fixed effects, and plot as a random effect (*SI Appendix, Fig. S5 and Table S8*).

Next, I explored whether high values for SLA plasticity and low values for IL plasticity are adaptive at large ΔPAR values, because these were the associations found in the garden experiment (Fig. 3C). It was not feasible to use a single fitness

measure such as lifetime seed production in the case of *V. chamaedrys* because it is a polycarpic perennial species with pronounced vegetative propagation. Therefore, I used four vital rates as proxies for fitness: 1) survival probability, 2) plant size, 3) flowering probability, and 4) seed production. In the analysis of survival probability, I included only those individuals that were alive during a check in April 2013, since mortality prior to April 2013 was likely due to transplant shock and not due to competition for light (given that the aboveground biomass of the invader dies back during winter). Lankau (45) used a similar approach. The other three vital rates were based on measurements obtained from surviving plants in June 2013.

Each vital rate was then used as the response variable in generalized linear mixed-effect models. As fixed effects, I used 1) plant size in November 2012, 2) genotype-specific trait plasticity (BLUPs) measured in the 2011 to 2013 garden experiment, 3) Δ PAR measured in June 2013 separately for each experimental plot, and 4) the trait plasticity \times Δ PAR interaction. Plot was included as a random effect. I ran two sets of models, one with SLA plasticity and the other with plasticity in IL. The binomial distribution was evaluated to analyze survival and flowering probability, and Poisson distribution was determined in the analysis of seed production. Normal distribution was then used in the analysis of plant size. Significance of fixed effects was tested by a likelihood-ratio test, and nonsignificant terms were removed to obtain minimum adequate models (SI Appendix, Table S9).

Modeling population growth with and without evolution of plasticity in shade-responsive traits. In this analysis, I explored whether adaptive evolutionary changes in SLA and IL plasticity can prevent the extinction of *V. chamaedrys* populations at very low Δ PAR values, that is, in light conditions typical for the early invasion stages (Fig. 2B). For this investigation, I first parameterized an IPM using vital rate functions obtained from the field experiment, while explicitly including trait plasticity (BLUPs) and its interaction with Δ PAR (SI Appendix, Table S10). In the second step, I estimated λ values by using IPMs for different Δ PAR values (for $n = 1,000$ values of Δ PAR from the uniform distribution on

the interval from 0 to 0.53, a range of observed Δ PAR values) while allowing or not allowing for the evolution of shade-responsive traits (according to a concept in SI Appendix, Fig. S6; see also ref. 46). I used four scenarios of evolution in trait plasticity that changed (or not) according to the Δ PAR values: 1) no evolution of plasticity (plasticity in both traits stays constant and corresponds to plasticity values of populations from uninvaded sites); 2) plasticity in SLA evolves according to Δ PAR but plasticity in IL stays constant; 3) plasticity in SLA stays constant but plasticity in IL evolves according to Δ PAR; and 4) plasticity in both traits evolves according to Δ PAR.

Relative contribution of changes in ecological and evolutionary factors to the variations in field-observed population growth. I used a method developed by Hairston et al. (47) to analyze the relative contributions of changes in ecological (log change in light availability) and evolutionary factors (evolution of shade-induced plasticity) to the variations in the field-observed population growth (log change in ramet density) of *V. chamaedrys*. I calculated the relative contributions of the two factors across three time intervals: from 0 (uninvaded state) to 11 y, from 11 to 28 y, and from 28 to 42 y of invasion (SI Appendix, Fig. S8 and Table S11).

Data Availability. The relevant data and IPM code can be accessed from Dataset S1.

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