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ORIGINAL PAPER

Contrasting levels of evolutionary potential in populations of the invasive plant *Polygonum cespitosum*

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Abstract The amount of quantitative genetic variation within an invasive species influences its ability to adapt to conditions in the new range and its long-term persistence. Consequently, this aspect of genetic diversity (or *evolutionary potential*) can be a key factor in the success of species invasions. Previous studies have compared the evolutionary potential of populations in introduced versus native ranges of invasive species, but to date no study has examined differences among introduced-range populations of such species in levels of quantitative genetic variation expressed in ecologically relevant environments. We assessed quantitative variation of fitness, life-history, and functional traits in six geographically separate introduced-range populations of the invasive annual *Polygonum cespitosum*, by comparing norms of reaction for a large sample of genotypes (16–19 per population) expressed in response to two glasshouse environments simulating contrasting habitats in this new range. Patterns of reaction norm diversity varied considerably among the 6 populations studied. Two populations showed very little quantitative genetic variation in both environments. In contrast, two other populations contained significant genetic variation for fitness and life-history traits in the form of genotypes with low performance in both habitats. Finally, two populations showed significant norm of reaction diversity in the form of cross-over interaction: genotypes that performed relatively well in one environment did poorly in the other. Differences among populations in potential selective response are likely to affect the dynamics and future spread of *P. cespitosum*, since specific populations will likely contribute differently to the invasion process. More generally, our results suggest that the evolutionary component of long-term invasion success may depend on population rather than on species-level processes.

Keywords Evolutionary potential · Quantitative variation · Invasion · Genotype × environment interaction · Reproductive traits

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Introduction

As a consequence of their introduction into different biogeographical regions, non-indigenous species are often subject to new abiotic and biotic conditions that

can impose novel selection pressures (Mooney and Cleland 2001; Sakai et al. 2001; Novak 2007; Prentis et al. 2008). The presence of quantitative genetic variation for functional and fitness traits within populations in a species' introduced range will contribute to its ability to adapt to such novel conditions through selective evolution, and therefore to successfully persist and spread. In other words, quantitative genetic variation for ecologically important traits is a key aspect of adaptive evolutionary potential of organisms in a new range (Fisher 1958; Sakai et al. 2001; Lee 2002; Byers 2005; Facon et al. 2008; Prentis et al. 2008; Matesanz et al. 2010; Miehls et al. 2011). Evolutionary potential of introduced-range populations will also influence a species' long-term persistence in a new range in the face of future environmental changes (Lee 2002; Parker et al. 2003; Dlugosch and Parker 2008a).

A key implication of this insight is that population-level differences in evolutionary potential can influence the long-term dynamics of an invasion (Huey et al. 2005; Lee and Gelembiuk 2008). If all populations in a species' introduced range possess similarly high levels of quantitative genetic variation, they will all be predicted to contribute to the invasive success of the species. However, if populations vary in adaptive evolutionary potential, the invasion trajectory may reflect the spread of a subset of evolutionarily labile populations rather than a moving front consisting equally of all populations (Lee and Gelembiuk 2008). Accordingly, comparisons among introduced-range populations of invasive species may provide important insights to invasion dynamics (Matesanz et al. 2012).

Despite the recognition that evolutionary change can be a key factor in the success of biological invasions, little is known about patterns of quantitative genetic variation in introduced-range populations of invasive species. Although numerous studies have assessed levels of neutral molecular variation in introduced taxa (reviewed in Dlugosch and Parker 2008a; DeWalt et al. 2011; Hardesty et al. 2012), information is comparatively scarce on quantitative genetic variation for functional and fitness traits expressed in ecologically relevant environments. Furthermore, most studies assessing such variation have aimed to compare differences in evolutionary potential between populations in the introduced versus native ranges of these taxa, considering populations within ranges to have equal genetic variances (e.g. Chen et al. 1991; Kaufman and Smouse 2001; Lavergne and Molofsky 2007; van Kleunen and Fischer 2008).

Here we present the first study comparing levels of ecologically relevant quantitative genetic variation among populations within the introduced range of an invasive species, using the well-studied Asian annual *Polygonum cespitosum*. *Polygonum* (s.l.) *cespitosum* Blume (=*Persicaria cespitosa*, Kim and Donoghue 2008) is a highly selfing species introduced from eastern Asia in the early 1900s (Paterson 2000) that has recently been catalogued as invasive in northeast North America (Mehrhoff et al. 2003). Previous studies have shown that introduced-range populations of this species can include individuals with high adaptive plasticity for functionally important traits, as well as genotype × environment variation for trait expression (Sultan 2001; Sultan et al. 2012). However, it is not yet known whether populations differ in this critical aspect of evolutionary potential. Investigations of neutral molecular variation in *P. cespitosum* have shown contrasting levels of microsatellite diversity in introduced-range populations as well as high population differentiation (Matesanz, Theiss, Holsinger and Sultan, in revision). These patterns of neutral genetic diversity are most likely the result of high selfing rates and limited seed dispersal ability as well as a history of multiple introductions. These factors may have also influenced patterns of quantitative genetic variation for adaptive traits among populations of the species.

Adaptive evolutionary potential can be assessed in populations of interest by comparing the reaction norms of a random sample of genotypes (or families) across a range of experimental treatments that mimic natural environmental variation (see Parker et al. 2003; Dlugosch and Parker 2008a; Facon et al. 2008). Using this type of quantitative genetics approach, it is possible to compare levels of genotypic and genotype × environment variation available to natural selection (Via and Lande 1985; Sultan 2007).

We studied a set of six populations that represent the current ecological distribution of the species in this part of its introduced range (see Matesanz et al. 2012). For each of these populations, we quantified genetic variation for a suite of life-history, morphological, physiological and reproductive traits expressed in response to two contrasting controlled environments: an open, dry treatment similar to high-light habitats in the introduced range and a shaded moist treatment

similar to the species' ancestral habitat both in Asia and initially in North America (Sultan et al. 1998). Evolutionary potential for adaptation to open, dry conditions is of particular interest because the frequency of such sites is predicted to increase in the future in this region, as summer droughts become more frequent due to climate change (Karl et al. 2009). Accordingly, quantitative genetic variation expressed in these two test environments is likely to be critical to the species' future success in northeast North America, where disturbed sites colonized by annuals vary strongly in light and moisture availability (Matesanz et al. 2012). We used standard quantitative genetics techniques to estimate genetic variance in each population for these ecologically meaningful traits, to address the following questions: (1) Do introduced-range populations of the invasive *P. cespitosum* show quantitative genetic variation (evolutionary potential) in response to simulated shade and open habitats? (2) If so, are populations similar or different in levels and patterns of variation? (3) What are the implications of these patterns of genetic diversity for future success of *P. cespitosum* in its introduced North American range?

Materials and methods

Experimental sample

Achenes were collected in October 2008 from 6 well-established populations at least 30 km apart, representing the species' current habitat range in northeastern North America (see Appendix S1, Electronic Supplementary Material for details on study populations). This sample included populations in forest understories where plants grew in the shade but received multiple daily sunflecks (GAY and JAM) as well as variable (temporally and spatially) populations where plants received full sun during part of the day or where shaded and full-sun microsites were present (ARM, HAR, WAD and WEI). All populations occurred in disturbed sites where both light and soil moisture varied within sites (see Fig. 1 in Matesanz et al. 2012). All populations occupied at least 100 m^2, with abundances of *Polygonum* reproductive individuals ranging from 50 to 175 plants/m^2 (i.e. all populations had at least 5,000 individuals, Appendix S1), and had similar levels of herbivory (percentage of leaf surface damaged by herbivores was lower than 10 % in all populations) and soil nutrients (Horgan-Kobelski, Matesanz and Sultan, in revision). Although the exact date of establishment of each population is unknown—which is the case for most introduced, rapidly spreading species—*P. cespitosum* was first reported in Connecticut and Massachusetts circa 1930 and occurred in shaded moist habitats (Blake 1932). Furthermore, field data and records of the Invasive Plant Atlas of New England (Sultan et al. 1998; Mehrhoff et al. 2003) indicate that the WEI and WAD populations have been established since at least 1992 (20 years).

In March 2009, achenes from 16 to 19 field individuals located $\geq$1 m apart along linear transects were collected from each population and grown to maturity in uniform, favorable glasshouse conditions, to produce inbred (selfed full-sib) genetic lines (hereafter *genotypes*) lacking maternal-environment differences (Griffith and Sultan 2012). Because *P. cespitosum* is a highly selfing species, full-siblings are highly homozygous and nearly identical (the inbreeding coefficient, F_{IS}, in these populations estimated from microsatellite markers ranges from 0.75 to 0.98; Matesanz, Theiss, Holsinger and Sultan, in revision).

Thirty-six mature achenes were collected from each of these inbred plants, air-dried, stored at 4 °C, and then stratified in distilled water for $\sim$4 week at 4 °C and sown into flats of moist vermiculite (8–10 June 2009). At the first true-leaf stage (5–7 July 2009), three replicate seedlings per genotype were randomly assigned to each of two experimental glasshouse environments (see below). The final sample included 609 plants (16–19 genotypes/population $\times$ 6 populations $\times$ 2 environments $\times$ 3 genotypic replicates/environment).

Experimental environments

Seedlings were individually transplanted into 1 l clay pots filled with a 1:1:1 mixture of medium sand (Quikrete Co., Atlanta, GA, USA), sterilized topsoil (Butler Construction, Portland, CT, USA) and Turface MVP fritted clay (Profile, Buffalo Grove, IL, USA), with 2.5 g per pot granular 15:8:12 NPK fertilizer (Agway, Middlefield, CT, USA). Seedlings received 75 % sun and were well-watered for 48 h to allow establishment, after which one replicate seedling per genotype was assigned to each treatment (Open/Dry and Understory/Moist) in each of three blocks

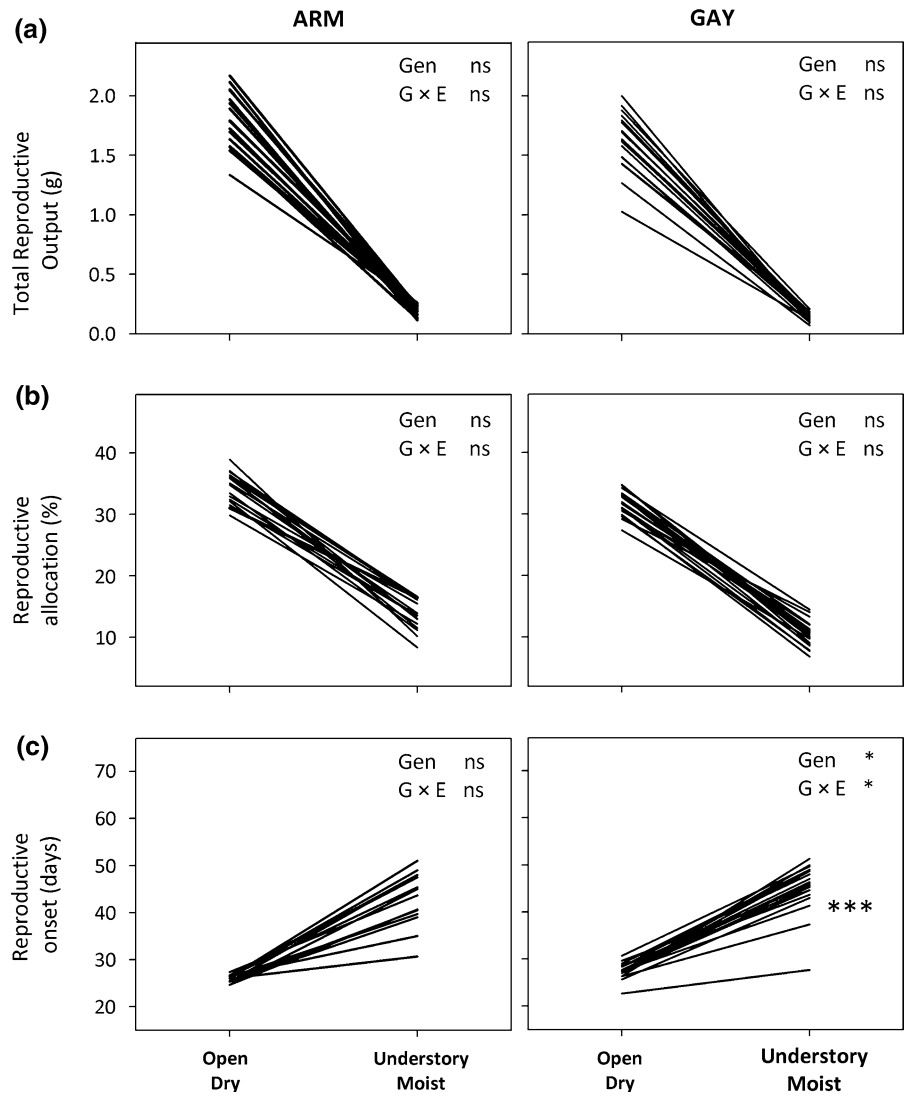

Fig. 1 Within-population genetic variation in fitness and life-history traits in Open/Dry versus Understory/Moist conditions for populations *ARM* and *GAY*. Norms of reaction for 16 and 19 genotypes per population, respectively of **a** total reproductive output, **b** reproductive allocation and **c** reproductive onset. Significance of the genotype (*Gen*) and genotype × environment interaction ($G \times E$) are shown. Environment was highly significant in all traits and populations ($P < 0.001$). Symbols show significant genetic variation in each environment. *ns* Not significant, $^{\dagger}P < 0.10$; $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$. See Appendix S2 and S3 for full results of the model

(contiguous glasshouse compartments containing both treatments) in a complete randomized block design (Zar 1999). These treatments were designed to mimic the extremes of the current species distribution in northeastern North America (Horgan-Kobelski, Matesanz and Sultan, in revision). Plants were grown in treatments for 10 weeks.

Plants in the Open/Dry environment received full sun (mean midday PAR $\sim$1,300 µmol m^{-2} s^{-1}). Understory/Moist plants were grown under metal frames covered with neutral 80 % shade cloth (PAK Unlimited Inc., GA, USA; mean midday PAR was c. 260 µmol m^{-2} s^{-1}) overlaid with green plastic filter strips (#138, Lee Filters, Burbank, CA, USA) to simulate canopy shade (Griffith and Sultan 2005). To mimic understory conditions, we created sunflecks to simulate the increase of direct solar radiation that occurs in forest understories when sunlight passes through openings in the canopy (Chazdon and Pearcy 1991; Valladares et al. 1997), by cutting equidistant 3.5 cm-diameter holes (one per pot) in the shade cloth. An extra row of holes was added along the frame edges to ensure that all pots received the same number of sunflecks. The metal frame was hung 35 cm above the bench and was situated so that the center of each pot received a $\sim$15 min-sunfleck at noon. This duration is typical of the shaded forest understories where *P. cespitosum* occurs (sunflecks lasting $\leq$15 min

represent ~90 % of all sunflecks occurring in these sites; Horgan-Kobelski, Matesanz and Sultan, in revision).

Soil moisture was maintained by automatic systems that delivered reverse osmosis-filtered water to one watering tube per pot (Chapin Watermatics, Watertown, NY, USA). Plants in the Open/Dry environment received 10–15 ml 3–4 times a day for a mean soil moisture of 50 % field capacity (9.23 ± 0.44 % by mass, based on 3 soil samples from individual pots at four time points during the experiment, N = 12). Understory/Moist plants received 15–20 ml 3–4 times a day, providing 100 % of field capacity (gravimetric soil moisture = 19.15 ± 1.19 %, N = 12).

Data collection

Physiological performance

Physiological measurements were taken on replicates of a subsample of 8 genotypes per population, for a total of 288 plants. Data were collected between 9 and 14 h on 6 comparable sunny days (12–19 August). On September 1, measurements were repeated for 28 plants identified as outliers in a preliminary data analysis. In situ *instantaneous photosynthetic rate* was measured on 1 new, fully-expanded leaf of a primary branch per plant using a Li-Cor 6400 infrared gas analyzer with red/blue LED light source and CO_2 mixer (LI-COR, Lincoln, NE, USA). Measurements were taken using a reference [CO_2] of 400 μmol CO_2 mol^{-1}, PPFD of 1,300 μmol m^{-2} s^{-1} in the Open/Dry environment and 300 μmol m^{-2} s^{-1} in the Understory/Moist environment, stomatal ratio of 0.7 (L. Nichols, unpublished data) and gas flow of 500 μmol s^{-1}. All plants were watered 30 min before measuring. Relative humidity was kept constant and close to ambient conditions (humidity range: 45–65 %); air temperature ranged from 30 to 38 °C. Measurements were logged only when the stability criteria were met (LI-COR 6400 User's manual).

Allocation and morphology

After 10 week in treatment (September 17–22), aboveground tissues of each plant were harvested, oven-dried (at 100 °C for 1 h and then 65 °C for ≥48 h) and weighed. Three non-senescent leaves from 1 primary branch per plant were scanned on an LI-3100 leaf area meter (LI-COR, Lincoln, NE, USA), oven-dried, and weighed to determine *specific leaf area* (SLA, leaf area/leaf biomass). Root systems were stored at 4 °C before being manually washed, oven-dried and weighed. Plant biomass was calculated as the sum of leaf, stem and root biomass.

Reproductive traits

Reproductive onset for each plant (date of first flowering, defined as the first day on which the petaloid sepals of at least a single flower were visible) was determined through a daily census. Mature achenes were collected weekly during week 5–10 in treatment. At final harvest (September 17–22), all remaining mature and immature achenes, flowers and reproductive support tissue were harvested. Achenes were air-dried for ≥5 days and weighed. *Total reproductive output* was calculated as the sum of the early maturing achenes plus all reproductive material collected at harvest. *Reproductive allocation* was calculated as (total reproductive output/plant biomass) × 100 %.

The measured traits have repeatedly been shown to be of critical importance in plant response to moisture- and light-limited conditions such as those imposed by our experimental treatments (see e.g. Grime 1977; Sultan and Bazzaz 1993a, b; Matesanz et al. 2012). Furthermore, a previous study showed that these traits were associated with fitness both in Understory/Moist as well as Open/Dry conditions in a sample of introduced-range *P. cespitosum* populations (Matesanz et al. 2012).

Data analyses

Mixed model ANOVA was used to test for the (fixed) main effects of environment (E) and block, the (random) main effect of genotype (G), and genotype by environment interaction (G × E). A significant main effect of genotype indicates that, on average, genotypes differ from each other, i.e. genetic variation for the trait; a significant effect of environment indicates plasticity for the trait; and a significant G × E interaction indicates that differences among genotypes are not consistent from one environment to another (i.e. genetic variation for plasticity). These models were repeated with restricted maximum likelihood (REML) mixed model estimations, and very

similar results were obtained. Because our goal was to examine and compare the responses of genotypes within each population to the experimental treatments, rather than comparing population mean differences in fitness and functional traits, the analyses were performed for each population separately. Finding significant G or G × E effects only in certain populations is interpreted as population differences in genetic variation and evolutionary potential.

Total reproductive output was (square-root) transformed to meet the assumptions of the model (Zar 1999). To minimize potential bias in the estimation of genotypic and genotype by environment effects associated with data transformation (Stanton and Thiede 2005), the analyses were performed using absolute (untransformed) fitness, square-rooted transformed fitness and relative fitness (calculated by dividing a genotype's fitness value by the mean fitness of all genotypes in each environment). Results for the three sets of analyses were virtually identical so only results for transformed data are shown. Pearson's correlation coefficients were calculated between genotypic-mean total reproductive output values in the two environments.

A second set of population-level analyses were performed within each environment, to test for the (random) effect of genotype (and the fixed effect of block). When significant genetic variation was detected within an environment, we used post hoc comparisons (linear contrasts) to test for differences among genotype(s) that appeared to be responding differently (Hill and Lewicki 2005). The goal of these contrasts was not to test any a priori hypotheses about specific genotypes, but only to determine whether their apparent differences in response across treatments were statistically robust (Baguley 2012; see Sultan and Bazzaz 1993a, b for a similar approach). That is, the contrasts simply clarify the data distribution and are not consulted as hypothesis tests. This approach is preferable to post hoc comparison of all possible pairs of genotypes because such mass post hoc testing can inflate type I error rates (Zar 1999).

To provide an index of genetically-based variance in each population, we additionally examined the proportion of phenotypic variance attributed to differences among genotypes within each environment and population, as Variance $_{GENOTYPE}$/Total Phenotypic Variance (see Conner 2003; Parker et al. 2003; Lavergne and Molofsky 2007; Dlugosch and Parker 2008b; Facon et al. 2008 for other studies using the same metrics). Variance components were estimated using REML. Significance of variance components were tested by likelihood ratio tests, by comparing the full model (including fixed and random factors) with the reduced model (dropping the random factor; see van Kleunen et al. 2002; Holland et al. 2003; Colautti et al. 2010).

REML analyses were performed in Proc. Mixed, SAS 9.2 (SAS Institute, Cary, NC, USA), and likelihood ratios were computed using library nlme in R (Pinheiro et al. 2012). All other analyses (mixed ANOVAs, linear contrasts and correlations) were performed in Statistica 8 (Tulsa, OK, USA).

Results

Although genotypes in all populations showed pronounced fitness and functional plasticity in response to contrasting light and moisture conditions (Environment $P \leq 0.001$ for all traits and populations, Appendix S2), the 6 populations exhibited strikingly different patterns of quantitative genetic variation for these traits (Appendix S2 and S3; Figs. 1, 2, 3, 4). The populations fell into three general types of pattern, described in detail below. Note that populations that shared a given pattern were not geographically the closest (average distance between populations sharing similar patterns: 80 km, minimum distance between populations: 30 km) nor did they occur in environmentally-similar sites (Appendix S1).

Low quantitative genetic variation: ARM and GAY populations

These two populations lacked significant genetic variation for fitness and functional traits, i.e. genotypes within each population showed similar patterns of response to the two experimental environments (Appendix S2, ns effects of Genotype and G × E interaction; Figs. 1, 4), with the single exception of reproductive onset within the UM environment in the GAY population (Fig. 1, right). Accordingly, the percentage of phenotypic variance explained by differences among genotypes was not significantly different from zero for all traits and environments (with the same one exception; Table 1). Genotype-mean total reproductive output was not correlated between environments in either population ($r =$

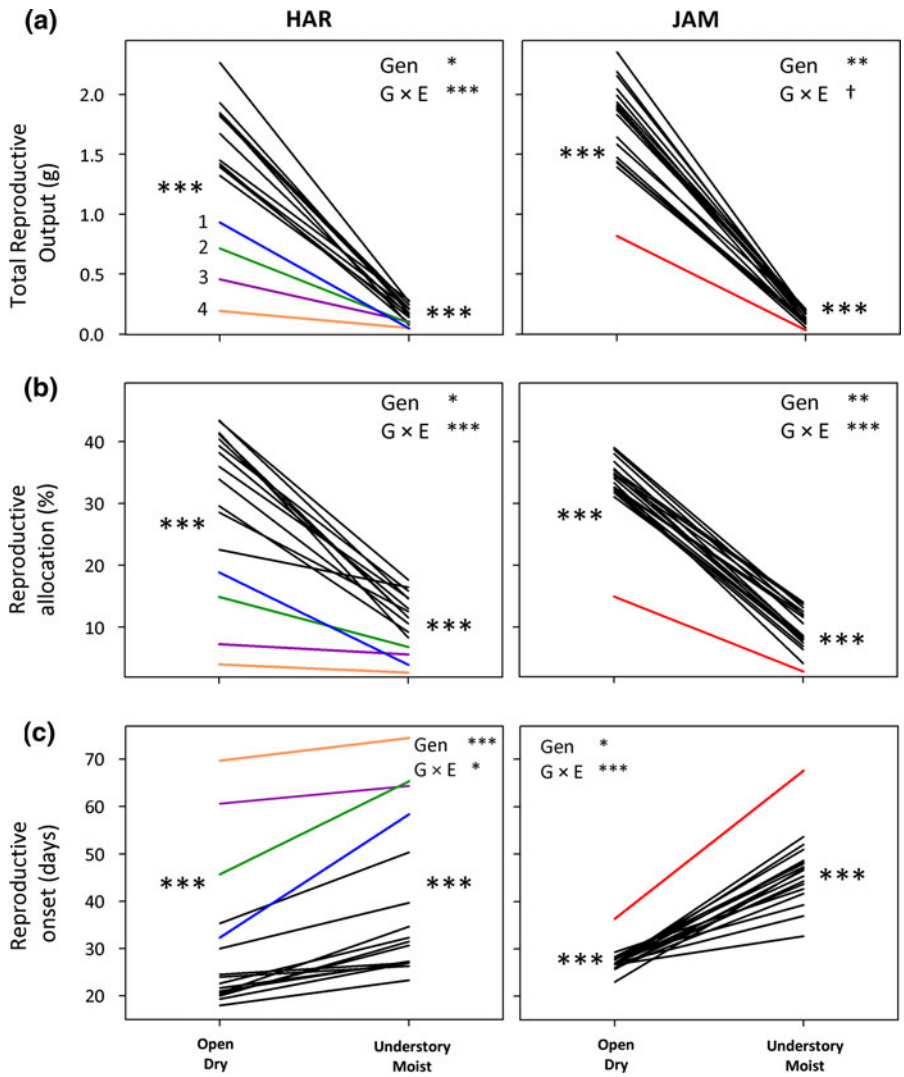

Fig. 2 Within-population genetic variation in fitness and life-history traits in Open/Dry versus Understory/Moist conditions for populations *HAR* and *JAM*. Norms of reaction for 17 and 19 genotypes per population, respectively of **a** total reproductive output, **b** reproductive allocation and **c** reproductive onset. Significance of the genotype (*Gen*) and genotype × environment interaction ($G \times E$) are shown. Environment was highly significant in all traits and populations ($P < 0.001$). Symbols show significant genetic variation in each environment. *ns* Not significant, $^{\dagger}P < 0.10$; $^{*}P < 0.05$; $^{**}P < 0.01$;$^{***}P < 0.001$. Genotypes 1–4 (in *color*) are significantly different from all other genotypes in both environments (see "Results"). See Appendix S2 and S3 for full results of the model. (Color figure online)

-0.23, $P = 0.39$ and $r = 0.25$, $P = 0.31$ for ARM and GAY, respectively).

High genetic variance due to consistent genotypic differences: HAR and JAM populations

These populations showed significant Genotype and G × E variation for total reproductive output, reproductive allocation and reproductive onset (Appendix S2), with significant differences among genotypes within both Open/Dry and Understory/Moist conditions (Appendix S3; Fig. 2). These among-genotype differences explained a large proportion of the total phenotypic variance (62–94 and 45–75 % for HAR and JAM, respectively, Table 1). There was also significant genetic variation in the HAR population (but not in JAM) for Specific Leaf Area (SLA) in the UM environment, and for photosynthetic rate in the OD treatment (Fig. 4; Appendix S3). Genotype-mean total reproductive output was positively correlated between the Open/Dry and Understory/Moist environments in both populations ($r = 0.66$, $P = 0.005$ and $r = 0.67$, $P = 0.002$ for HAR and JAM, respectively), i.e. genotypes with relatively high or low reproductive output in one environment also had relatively high or low fitness in the other environment.

In HAR, the highly significant main effect of genotype for all 3 reproductive traits reflected the relatively low trait values in both environments of 4

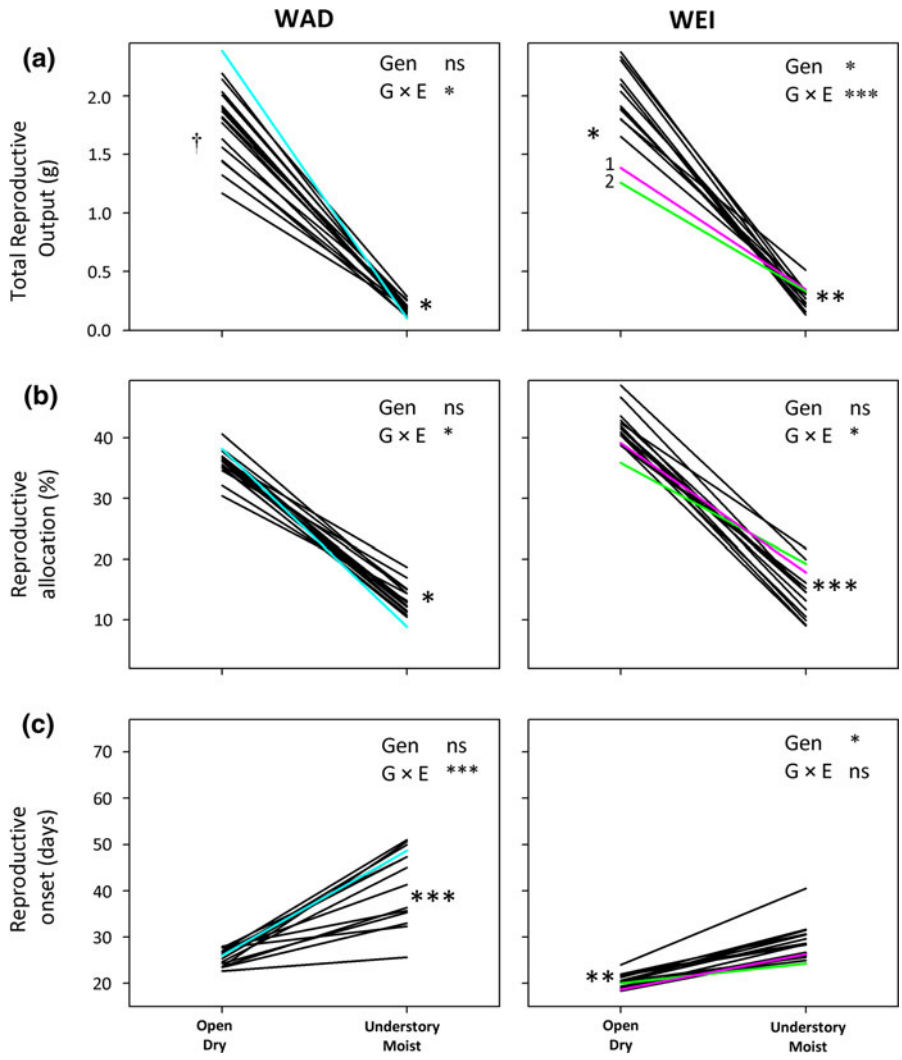

Fig. 3 Within-population genetic variation in fitness and life-history traits in Open/Dry versus Understory/Moist conditions for populations *WAD* and *WEI*. Norms of reaction for 17 and 16 genotypes per population, respectively of **a** total reproductive output, **b** reproductive allocation and **c** reproductive onset. Significance of the genotype (*Gen*) and genotype × environment interaction ($G \times E$) are shown. Environment was highly significant in all traits and populations ($P < 0.001$). Genotypes *highlighted in color* show cross-over interactions between treatments. Symbols show significant genetic variation in each environment. *ns* Not significant, $^{\dagger}P < 0.10$; $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$. See Appendix S2 and S3 for full results of the model. (Color figure online)

genotypes which showed consistently lower reproductive output (~66 and 60 % lower reproductive output in the O/D and U/M environments, respectively), lower reproductive allocation (~150 % lower in both environments), and delayed reproductive onset (by an average of 35 and 29 days, genotypes 1–4 in Fig. 2 left). These genotypes each differed significantly from other genotypes within both environments (linear contrasts for each of four genotypes vs. all other genotypes, $P < 0.013$, $P < 0.039$ and $P < 0.001$ for reproductive output, reproductive allocation and reproductive onset).

Similarly, the significant genetic variation for fitness traits in the JAM population reflected the low reproductive output (54–75 % in the OD and UM environment, respectively), low allocation to reproduction (140–156 %) and delayed reproductive onset (by 9.5–22.5 days) of one genotype in both environments (highlighted in Fig. 2 right; linear contrasts, $P < 0.001$ for all traits). Because these consistently low-performing genotypes expressed less fitness plasticity in response to high-light conditions (Open/Dry environment), the G × E term as well as the average effect of Genotype were significant (Fig. 2).

High genetic variance due to crossover interaction between environments: WAD and WEI populations

These populations showed significant G × E interaction for all 3 reproductive traits (Appendix S2; Fig. 3), except for reproductive onset in WEI. Genotype-mean

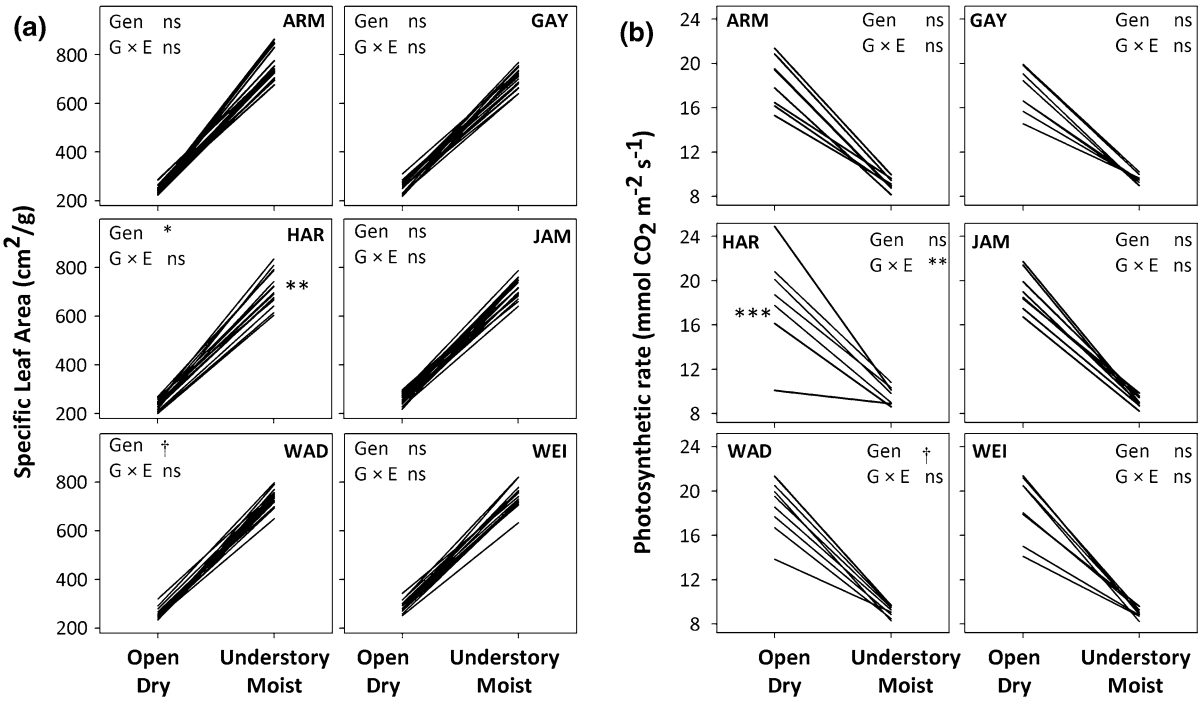

Fig. 4 Within-population genetic variation in **a** specific leaf area and **b** photosynthetic rate in Open/Dry versus Understory/Moist conditions for all populations. Norms of reaction for 16–19 genotypes per population (8 genotypes for photosynthetic rate). Environment was highly significant in all traits and populations ($P < 0.001$). Genotypic and $G \times E$ effects are only significant (or marginally) in two instances. *ns* Not significant, † $P < 0.10$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$. See Appendix S2 and S3 for full results of the model

total reproductive output was not correlated between environments in either population ($r = -0.09$, $P = 0.74$ and $r = -0.28$, $P = 0.29$ for WAD and WEI, respectively). No significant variation for SLA or photosynthetic rate was found in either population (Appendix S2).

In the WAD population, there was significant genetic variation for the three reproductive traits within the Understory/Moist environment (Appendix S3), but in the Open/Dry treatment the genotype effect was non-significant for reproductive allocation and onset (Fig. 3, Appendix S3). Accordingly, the amount of variance explained by genotypic differences of reproductive traits was higher in the Understory/Moist treatment than in the Open/Dry treatment (Table 1). In this population, the significant G × E interaction effect on fitness traits reflected one genotype (highlighted in Fig. 3 left) that had the highest fitness and 2nd-highest reproductive allocation in the Open/Dry environment and the lowest fitness and allocation in the Understory/Moist environment (linear contrasts for total reproductive output vs. all other genotypes, $P = 0.014$ and $P = 0.018$, respectively; ns effect of G × E after removing this genotype from the analysis).

In the WEI population there was significant genetic variation for total reproductive output within both environments (Appendix S3; Fig. 3), as well as G × E interaction reflecting changes in the rank order of certain genotypes (highlighted in Fig. 3 right). The two genotypes with the lowest reproductive output in the Open/Dry environment (genotypes 1–2 in Fig. 3, right) had (marginally significantly) higher reproductive output in the Understory/Moist environment (linear contrasts vs. all other genotypes, $0.016 < P < 0.17$). A similar pattern was found for reproductive allocation in these two genotypes (Fig. 3).

Discussion

Although *P. cespitosum* is a highly inbreeding species, our study of 6 North American populations revealed significant quantitative genetic variation in fitness and

Table 1 Percentage of total phenotypic variance (%PhVa) attributed to differences among genotypes for fitness and functional traits within Open/Dry (top panel) and Understory/Moist conditions (bottom panel) in 6 introduced-range populations of *P. cespitosum*

	Total repro. output		Repro. allocation		Repro. onset		SLA		Photo. rate	
	% PhVa	χ^2	% PhVa	χ^2	% PhVa	χ^2	% PhVa	χ^2	% PhVa	χ^2
Open/Dry environment										
ARM	0.9	0.004ns	6.4	0.140ns	0.0	0.000ns	0.0	0.000ns	42.1	3.155^{+}
GAY	2.5	0.0301ns	0.0	0.0000ns	3.4	0.061ns	15.3	1.081ns	0.0	0.000ns
HAR	73.6	25.296***	88.6	43.127***	93.6	62.194***	0.0	0.000ns	76.9	9.707**
JAM	45.0	9.905**	75.2	33.224***	66.5	23.934***	9.8	0.478ns	16.1	0.494ns
WAD	23.8	2.733^{+}	3.2	0.046ns	16.1	1.056ns	0.0	0.000ns	26.4	0.001ns
WEI	29.8	2.999^{+}	21.3	1.767ns	45.0	6.029*	22.1	1.892ns	0.0	0.000ns
Understory/Moist environment										
ARM	4.6	0.090ns	8.5	0.304ns	23.2	2.134ns	0.0	0.000ns	14.1	0.389ns
GAY	0.0	0.000ns	5.4	0.151ns	54.0	13.005***	0.0	0.000ns	0.0	0.000ns
HAR	61.3	15.760***	80.0	32.728***	84.0	34.223***	38.9	5.991*	10.7	0.226ns
JAM	51.6	13.345***	58.9	18.009***	56.6	16.415***	1.8	0.017ns	0.0	0.000ns
WAD	31.4	4.075*	29.7	3.852*	54.8	13.573***	0.0	0.000ns	0.0	0.000ns
WEI	45.1	6.315*	54.5	10.587**	15.2	0.523ns	4.1	0.045ns	0.7	0.001ns

Variance components were estimated by restricted maximum likelihood (REML) and tested by likelihood ratio test (χ^2 and P values shown)

ns Not significant

$^{+}$ $P < 0.1$; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

life-history traits. Genetic variation was expressed in each of two experimental treatments that simulated contrasting habitats in the species' introduced North American range: moist understory and open, dry conditions. These results indicate that this non-native species has substantial evolutionary potential to adapt to variation in light and moisture conditions, which may contribute to its future persistence and spread in this new range (Sakai et al. 2001; Novak 2007). Genetic variation for reproductive timing, allocation and total output is particularly notable because these traits contribute directly to propagule pressure, an important factor in invasion success (Lockwood et al. 2005).

However, populations sampled from the species' introduced range differed in levels and patterns of quantitative genetic variation in the two contrasting environments. A sample of just six introduced-range populations revealed three different patterns of genetic diversity. To our knowledge, this is the first study to directly document differences in adaptive evolutionary potential among introduced-range populations of an invasive species. For non-invasive taxa, population differences in quantitative variation have been observed in both plants and animals (see e. g. Black-Samuelsson and Andersson 1997; Donohue et al. 2001; Gomez-Mestre and Tejedo 2004; Knopp et al. 2007). Although, in some cases, these differences may reflect the past action of local selection pressures, they are generally considered to result from population-level evolutionary factors such as founder effects, dispersal history, and inbreeding (i.e., population size and structure) that shape adaptive potential. For instance, the classic study of Al-Hiyaly et al. (1988, 1993) examined populations of a native grass growing in similarly zinc-contaminated soils, and found that these populations showed contrasting zinc tolerance. They concluded that different levels of genetic variation among founding populations resulted in different potential to evolve zinc resistance despite similar selection pressures in the various sites. Our results are thus consistent with evolutionary studies in non-invasive taxa that show how the founding history and structure of local populations can lead to differences in their potential for subsequent adaptive change.

In two of the *P. cespitosum* populations, genotypes shared largely uniform norms of reaction: for most

traits, genotypes in these populations did not differ significantly within either environment. Lack of genetic variation in traits of adaptive significance indicates that further evolution of these introduced-range populations in response to light and moisture variation may be limited (Byers 2005). Similarly, Parker et al. (2003) found extremely low among-family variation for morphological and physiological traits in populations of the invasive weed *Verbascum thapsus*.

Conversely, we found significant, consistent among-genotype variation for fitness and life-history traits in a second pair of (geographically distinct) populations, consistent with other studies reporting overall high evolutionary potential in the introduced range of invasive taxa (e.g. Lavergne and Molofsky 2007; Facon et al. 2008; Miehls et al. 2011). In these populations, certain genotypes ranked either higher or lower than others in both Open/Dry and Understory/Moist conditions. This pattern of consistent genotypic performance differences across contrasting environments provides potential for the evolution of generalist, high-performance genotypes (Falconer and Mackay 1996; Blows and Hoffmann 2005) that may fuel a species' invasive spread across diverse habitats (Matesanz and Sultan in review; Le Roux et al. 2007). Evolution can be constrained if there are genetic correlations among traits, even in the presence of significant genetic variation for the traits (Blows and Hoffmann 2005; Colautti et al. 2010). In our study, genetic correlations are not likely to limit the potential for evolution in these two populations. A previous study of *P. cespitosum* populations grown in the same experimental treatments showed that fitness was positively associated with high allocation to reproductive tissues and early flowering in both environments (Matesanz et al. 2012). In the HAR and JAM populations, correlations among traits showed that there is genetic variation for the combination of traits that would allow selection to simultaneously improve both traits (significant negative correlation between reproductive allocation and reproductive onset in both populations and environments; data not shown).

The third pattern of quantitative genetic variation exemplified *crossover interactions* (Baker 1988), in which genotypes achieving high fitness in one environment had relatively low fitness in the contrasting environment. This pattern of genetic variation (identified by significant G × E interaction in the absence of significant genotype main effects) can have important implications for selection. When the expression of genetic variation is environmentally dependent, the availability of genetic variation to selection will depend on both the patterns of diversity among genotypes and the distribution of environments (Sultan and Bazzaz 1993a, b; Falconer and Mackay 1996; Byers 2005; Kingsolver et al. 2007; Sultan 2007). If norms of reaction cross between environments that occur within a given population (i.e. with fine-grained temporal or spatial variation), diverse genotypes may persist (Via and Lande 1985; Gillespie and Turelli 1989; Sultan 2007), since genotypes do not have relatively high or low fitness in all conditions that occur (Sultan and Bazzaz 1993a, b; Blows and Hoffmann 2005; Byers 2005). Alternatively, if each population encounters only a single type of environment, this pattern of crossover variation can lead to the evolution of specialized local ecotypes, as certain genotypes will be selectively favored in each environment. Furthermore, interactions of the crossover type suggest that performance in diverse environments is decoupled, such that new adaptive norms of reaction could evolve that may maximize fitness in contrasting conditions (Via and Lande 1985).

As is the case for non-invasive species, contrasting patterns of quantitative genetic variation in the study populations may result from several non-mutually exclusive factors. Lack of significant genetic variation in specific populations may be due to founder effects and/or previous selection in these sites (Lee 2002; Blows and Hoffmann 2005; Le Roux et al. 2007; Prentis et al. 2008). Conversely, in populations with high quantitative genetic variation, the presence of genotypes expressing low fitness in one or both experimental environments indicates that similar environmental conditions may have occurred too infrequently in their respective sites, or that the populations had been established too recently for them to have been eliminated by selection (Ghalambor et al. 2007; Griffith and Sultan 2012 and references therein). Multiple introductions can also lead to high variation in populations of invasive animal and plant taxa (e.g. Ellstrand and Elam 1993; Kolbe et al. 2004; Maron et al. 2004; Lavergne and Molofsky 2007; Facon et al. 2008). For example, the HAR population showed relatively high expected heterozygosity ($H_e = 0.371$) and admixture of different genetic clusters (based on Bayesian assignment tests), suggesting that this

population may have resulted from several introductions (Matesanz, Theiss, Holsinger and Sultan, in revision).

Interestingly, populations that shared a given pattern of quantitative variation were not the closest, they did not occur in similar habitat types nor did they have similar environmental conditions. It may be possible that the expression of genetic variation in natural conditions is affected by variation in environmental factors other than light and soil moisture. However, field data from these populations indicate that light and soil moisture are the best predictors of plant performance in natural conditions (Horgan-Kobelski, Matesanz and Sultan, in revision). Although population-specific patterns of genetic variation may be altered by gene flow among populations (Etterson and Shaw 2001; Lavergne and Molofsky 2007), in this system gene flow is likely to be limited as the species is highly inbred and has low, gravity-based seed dispersal.

Although the precise causes of among-population differences cannot be determined with certainty, identifying such differences provides important insights to invasion dynamics. Differences in genetic variation and evolutionary potential among populations are likely to affect the dynamics of introduced species and shape their invasion trajectory, since specific populations will likely contribute differently to the invasion process. The contribution of specific populations to the spread of the species will depend not only on the presence of such quantitative genetic variation but also on the nature of the genotypes present in the populations and the likelihood of encountering different environments. Populations with no genetic variation will likely contribute differently to the invasion process depending on whether they consist of high- or low-performing genotypes in specific environments (e.g. ARM vs. GAY populations). For example, the ARM population contains genotypes that are able to perform better in open, dry conditions than those present in the GAY population. High performance in such conditions is particularly relevant for invasion potential, since *P. cespitosum* has recently expanded its ecological range from shade, moist environments to more commonly inhabit sites with increased mean light availability and potential moisture deficits (Horgan-Kobelski, Matesanz and Sultan, in revision). In these cases, a population's lack of genetic variation may not constrain its invasion potential.

The existence of substantial among-population differences in evolutionary potential suggests that invasion success may depend to some extent on the ability of specific populations to adapt to habitats encountered in the new range, rather than on the species-level properties that are generally studied. Studies comparing quantitative genetic variation between the native and introduced range often assume equal variation within populations in each range (Kaufman and Smouse 2001; Chen et al. 2006; Lavergne and Molofsky 2007). Instead, our data indicate that among-population differences should be considered in predicting an introduced species' potential to adapt to a new range.

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