

Trait differences between and within ranges of an invasive legume species

Carmen Hoffbeck (✉ carmen.hoffbeck.50@my.csun.edu)

California State University Northridge <https://orcid.org/0000-0002-0685-8505>

Casey P terHorst

California State University Northridge

Research Article

Keywords: evolution, genetic variation, life history traits, Medicago polymorpha, rapid evolution

Posted Date: January 12th, 2022

DOI: <https://doi.org/10.21203/rs.3.rs-1197357/v1>

License:   This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

1 Trait differences between and within ranges of an invasive legume species

2

3 Carmen Hoffbeck¹

4 Casey P. terHorst¹

5 ¹Department of Biology, California State University, Northridge

6

7 Keywords: evolution, genetic variation, life history traits, *Medicago polymorpha*, rapid

8 evolution

9

10 **Abstract**

11 Novel ecological interactions can drive natural selection in non-native species and
12 trait evolution may increase the likelihood of invasion. We can gain insight into the potential
13 role of evolution in invasion success by comparing traits of successful individuals in the
14 invasive range with the traits of individuals from the native range in order to determine which
15 traits are most likely to allow species to overcome barriers to invasion. Here we used
16 *Medicago polymorpha*, a non-native legume species from the Mediterranean that has invaded
17 six continents around the world, to quantify differences in life history traits among genotypes
18 collected from the native and invasive range and grown in a common greenhouse
19 environment. We found significant differences in fruit and seed production and biomass
20 allocation between invasive and native range genotypes. Invasive genotypes had greater
21 fecundity, but invested more energy into belowground growth relative to native genotypes.
22 Beyond the variation between ranges, we found additional variation among genotypes within
23 each range in flowering phenology, total biomass, biomass allocation, and fecundity. We
24 found non-linear relationships between some traits and fitness that were much stronger for
25 plants from the invasive range. These trait differences between ranges suggest that stabilizing
26 selection on biomass, resource allocation, and flowering phenology imposed during or after
27 introduction of this species may increase invasion success.

28

29 **Introduction**

30 Novel environments represent a multitude of challenges to which invading species
31 must adapt, particularly when invaders are adapted to substantially different conditions in
32 their native range (Parker & Gilbert, 2007; Zenni & Nuñez, 2013; Hui et al., 2016). Despite
33 these challenges, potential invaders are often afforded clear advantages in new ecosystems,
34 including naïve competitors or other natural enemies who may not be adapted to harm novel
35 species and offer some release from antagonistic interactions relative to the native range
36 (Keane & Crawley, 2002; Lau & Strauss, 2005; Maron et al., 2014). Although some invaders
37 tend to invade habitats similar to the native habitat in which they evolved for long periods of
38 time, many invaders spread to a variety of novel habitats with a range of climates and
39 ecological interactions. The ability to spread to multiple habitat types depends on the ability
40 of a species to cope with a variety of environmental conditions. Depending on the species,
41 successful strategies can rely on phenotypic plasticity, selection on and evolution of inherited
42 traits, or both (Davidson et al., 2011; Palacio-López & Gianoli, 2011; Geng et al., 2016;
43 Marchini et al., 2019).

44 Though evolution is often considered over very long time periods, there is extensive
45 evidence that evolution often occurs on ecologically relevant time scales (Thompson, 1998;
46 Hairston et al., 2005; Schoener, 2011). For invasive species, evolution on ecological time
47 scales can be crucial for adaptation to a novel ecosystem (Maron et al., 2004; Lambrinos,
48 2004). In one example, invasive St. John's wort (*Hypericum perforatum*) displayed changes
49 in life history traits (size and fecundity) in response to environmental clines, indicating rapid
50 evolution to new environments (Maron et al., 2004). Similarly, high genetic variation and
51 repeated introductions of the invasive grass *Phalaris arundinacea* allowed for vegetative
52 colonization ability and greater phenotypic plasticity to evolve in the invasive range
53 (Lavergne & Molofsky, 2007). Depending on the mode of introduction of a novel species, the

54 invasion process may impose selection for greater dispersal, higher fecundity, larger size or
55 competitive ability, or other life history traits associated with invasion success (Jelbert et al.,
56 2015).

57 Trait variation within a species has been recognized as an important driver of many
58 ecological phenomenon, including niche selection and expansion, nutrient usage,
59 competition, and community structure (Hughes et al., 2008; Agashe & Bolnick, 2010). The
60 spectrum between r and K life history strategies (MacArthur & Wilson, 1967) is often
61 considered as differences among species, but is also relevant in considering differences
62 among individuals within a species (Bolnick et al., 2011). Differences in life history
63 strategies within a species may allow a species to fill more niche space – generalist species
64 can be composed of a population of specialist individuals (Bolnick et al., 2003; Agashe &
65 Bolnick, 2010). Genetic diversity is also important for rapid evolution, as such diversity is
66 required for natural selection to act upon. If high genetic diversity enables a population to be
67 respond to different types or strengths of species interactions and or utilize more resources
68 and expand the range of the niche, we might expect some genotypes in the species to
69 experience greater invasion success, or experience positive selection in a broader range of
70 ecosystems.

71 Life history strategies have been shown to be important targets of selection in
72 invasive species (Barrett et al., 2008). However, ecological pleiotropy or genetically-linked
73 traits suggest that selection on life history traits that promote invasion into a novel ecosystem,
74 must be considered against selection on linked traits or in response to multiple interactions.
75 For example, an increase in fecundity, which is typically positively related to invasion
76 success (Jelbert et al., 2015) may be selected for in an introduced population, but if increased
77 seed number is associated with decreased seed size, which decreases competitive ability, this
78 presents a trade-off in life history strategies. Adaptation to a new ecosystem requires

79 considering evolution of linked and correlated traits in the community context in which
80 invasion occurs (terHorst, Zee, et al., 2018).

81 *Medicago polymorpha*, commonly known as burr clover or burr medic, is an invasive
82 legume species that often competes with native species in the introduced range (Lau &
83 Strauss, 2005). *M. polymorpha* is endemic to the Mediterranean region, but has invaded six
84 continents, predominantly spreading from its use in agriculture as a nitrifying cover crop
85 (Porter & Simms, 2014). Understanding *M. polymorpha* and the traits that facilitate invasion
86 success can provide insight into invasive plant strategies more broadly, and is important for
87 limiting the spread of this invader into further habitats (Westbrooks, 1998). Previous research
88 has shown significant genetic variation in interactions between *M. polymorpha* and other
89 species, including competitors, herbivores, and mutualists (terHorst & Lau, 2015; Bayliss et
90 al., 2017; Getman-Pickering et al., 2018; terHorst, Wirth, et al., 2018). Such genetic
91 variation, as well as the ability to experimentally replicate genotypes, presents the
92 opportunity to compare genotypes of *M. polymorpha* that have been successful in the
93 invasive and native ranges. We can use such comparisons to examine any differences in life
94 history traits between genotypes that were able to successfully establish in novel habitats and
95 those genotypes successful in the native range. This can provide insight into potential
96 evolutionary shifts that may have taken place during or after the invasion process and may
97 have played a role in successful invasion by this species. We used a greenhouse experiment
98 to quantify differences in life history traits among and between genotypes from invasive and
99 native ranges to examine extant variation in this species and consider a possible role of
100 evolution in invasion success.

101

102 **Methods**

103 *Collection and rearing*

104 We haphazardly selected 10 accessions of *Medicago polymorpha* from the native and
 105 invasive ranges of *Medicago polymorpha* (Bullitta et al., 1994), from the USDA Germplasm
 106 Resource Information Network collection, for a total of 20 genotypes (Table 1). Because
 107 these genotypes have been grown in greenhouses for several generations after collection from
 108 their natural environment, maternal effects should play little role in determining traits. As
 109 these plants are primarily selfing, which should result in homozygous clones after a few
 110 generations, we refer to each accession as a genotype.

111

112 **Table 1.** USDA GRIN accession numbers for *Medicago polymorpha* with country of origin
 113 and range for each genotype.

Genotype	Country of Origin	Range	Genotype	Country of Origin	Range
260270	Ethiopia	Invasive	170546	Turkey	Native
201206	Australia	Invasive	170547	Turkey	Native
197346	Russia	Invasive	170548	Turkey	Native
186329	Australia	Invasive	197345	Morocco	Native
186963	Uruguay	Invasive	197344	Malta	Native
186964	Uruguay	Invasive	198964	Cyprus	Native
197336	Australia	Invasive	206380	Cyprus	Native
197338	United States	Invasive	206695	Turkey	Native
197859	Uruguay	Invasive	239870	Morocco	Native
202809	Peru	Invasive	205428	Turkey	Native

114

115 We physically scarified seeds from each genotype and then planted them in 164 mL
 116 cone-tainers (Stuewe and Sons, Tangent, OR, USA) filled with low-nutrient soil (Sun-Gro
 117 Sunshine mix #5). Plants were grown in the greenhouse at California State University,
 118 Northridge. We planted three seeds in each cone-tainer and weeded the number of seedlings

119 down to one per cone-tainer. This resulted in 240 total replicate cone-tainers (2 ranges x 10
120 genotypes x 12 replicates). Cone-tainers were randomly placed into racks and re-randomized
121 periodically to ensure that no genotypes received uniform exposure to any conditions. Cone-
122 tainers were watered every day for the first week and then every other day for the remainder
123 of the experiment.

124 Halfway through the experiment, plants were fertilized once with Grow More
125 fertilizer (Grow More Inc, Gardena, CA, USA; 20N:20P:20K) at ¼ strength. During the
126 experiment, we recorded the date on which each plant produced its first flower. We collected
127 each fruit produced by each plant after fruits had ripened and quantified the number and
128 weight of fruits per plant, as well as the number and weight of total seeds per plant. When no
129 fruits or flowers remained on the plants, we harvested each plant, which occurred between
130 140-169 days after planting. Plants that had not produced any flowers by this time were also
131 harvested and noted to have not reproduced. Once harvested, we rinsed the soil from the roots
132 of plants, separated aboveground and belowground portions, and placed them in a drying
133 oven at 50°C for two days. We then measured the total dry weight and the
134 above:belowground biomass ratio.

135

136 *Data Analysis*

137 We used generalized linear mixed models to test the fixed effects of range (native or
138 invasive) and genotype (random effect nested within range) on several dependent variables
139 in R (version 4.0.3). Our dependent variables were: survival to reproduction or the end of the
140 experiment, time to first flower, fruit number, total fruit weight, seed number, total seed
141 weight, total biomass, and above:belowground biomass ratio. For all variables except
142 survival, data analyses were restricted to plants that survived. We tested each variable for
143 normality and homogeneity of variances, and when possible, transformed the data to meet

144 these assumptions and fit models using ‘lmer’ in the lme4 package. Biomass ratio was
145 transformed with a natural log transformation and total biomass by an inverse square root
146 transformation. When variables could not be transformed to meet assumptions, we fit
147 alternative error distributions using ‘glmer’. Survival was analyzed using a binomial error
148 distribution and fruit and seed weight were analyzed with log-normal error distributions. Fruit
149 and seed number were analyzed using a zero-inflated distribution in the ‘zeroinfl’ package.
150 We tested the significance of fixed effects using ‘Anova’ in the car package, with Type III
151 sums of squares, and tested random effects using Likelihood Ratio Tests. To examine
152 relationships between pairs of dependent variables, we used similar models, but included
153 each dependent variable as a covariate in the models of every other dependent variable. Using
154 these models, we tested the linear and quadratic components of each pairwise relationship,
155 and examined whether these relationships differed between invasive and native ranges.

156

157 **Results**

158 *Survival rate and time to flower*

159 The survival of plants in the experiment was 67.5% across all ranges and genotypes.
160 There was no significant effect of range on survival ($X^2 = 0.485$, $df = 1$, $P = 0.486$), though
161 there were significant differences among plant genotypes independent of range ($X^2 = 106$, df
162 $= 1$, $P < 0.001$). One invasive genotype (186963 from Uruguay) experienced a 100%
163 mortality rate, but six invasive genotypes and eight native genotypes experienced 100%
164 survival rates, with the remaining five genotypes showing intermediate survival. There was a
165 marginally significant difference in flowering time between ranges ($F_{1,14} = 3.30$, $P = 0.089$),
166 with invasive genotypes tending to flower earlier than native genotypes. However, there was
167 also significant variation among genotypes within each range ($X^2 = 84.6$, $df = 1$, $P < 0.001$).
168 There was more variation in flowering times among native genotypes—one native (239870

169 from Morocco) and one invasive genotype (260270 from Ethiopia) flowered earlier than any
170 others, but two native genotypes were the last to flower (Fig. 1).

171
172 *Mean total biomass and above:belowground biomass ratio*

173 There were marginally significant differences in total biomass between native and
174 invasive range individuals ($F_{1,15} = 4.21$, $P = 0.058$), and significant differences among
175 genotypes within ranges ($X^2 = 22.9$, $df = 1$, $P < 0.001$). Despite significant variation within
176 ranges, invasive genotypes tended to be smaller than native genotypes (Fig. 2A). Similarly,
177 above:belowground biomass ratio was marginally significantly different between native and
178 invasive ranges ($F_{1,160} = 3.17$, $P = 0.07$). The difference in total biomass between ranges was
179 due largely due to differences in aboveground biomass, with invasive genotypes investing
180 relatively less in aboveground than belowground biomass compared to native range
181 genotypes (Fig. 2B). Unlike with total biomass, we observed no significant difference among
182 genotypes with ranges ($F_{1,11} = 0.385$, $P = 0.548$).

183

184 *Fruit and seed production*

185 Both fruit and seed number were significantly different between native and invasive
186 ranges (Figs. 3, S1), with invasive genotypes producing more fruits ($X^2 = 10.2$, $df = 1$, $P =$
187 0.001) and more seeds ($X^2 = 73.6$, $df = 1$, $P < 0.001$). In addition to differences between
188 ranges, genotypes also varied significantly in both fruit ($X^2 = 11.0$, $df = 1$, $P < 0.001$) and
189 seed number ($X^2 = 76.4$, $df = 1$, $P < 0.001$). However, we found no significant differences in
190 total fruit or seed weights between ranges (fruit weight: $X^2 = 1.14$, $df = 1$, $P = 0.286$; seed
191 weight: $X^2 = 1.97$, $df = 1$, $P = 0.161$, Fig. S2). The disparity between quantity and quality of
192 fruits and seeds does not seem to be due to a trade-off between number and size, as there is
193 significant positive correlation between fruit number and fruit weight ($F_{1,160} = 323$, $P <$
194 0.001) and between seed number and seed weight ($F_{1,160} = 1317$, $P < 0.001$). Rather, there

195 was more variability in weight than in number, resulting in decreased power to detect any
196 difference between ranges. There was significant variation among genotypes in fruit weight
197 and seed weight, beyond that explained by range ($X^2 = 72.351$, $df = 1$, $P < 0.001$; Fig. 3).

198

199 *Relationships among life history traits*

200 We found three cases in which the relationships between traits and fitness differed
201 between the native and invasive range. There was a non-linear relationship between seed
202 number and total biomass ($F_{1,156} = 10.6$, $P = 0.001$), but the strength of that relationship
203 differed among plants from the native and invasive ranges ($F_{1,156} = 53.2$, $P < 0.001$). Plants
204 from the invasive range with intermediate biomass produced more seeds than plants with
205 high or low biomass, but there was no such relationship for plants from the native range (Fig.
206 4A). Similarly, there was a significant non-linear relationship between seed number and
207 above:belowground biomass ratio ($F_{1,156} = 11.9$, $P < 0.001$) that was stronger for plants from
208 the invasive range ($F_{1,156} = 66.7$, $P < 0.001$). Invasive range plants with intermediate
209 above:belowground ratios tended to produce the most seeds, but this pattern was much
210 weaker for genotypes from the native range (Fig. 4B). Finally, there was a significant non-
211 linear relationship between seed number and flowering phenology, but the relationship
212 differed depending on range ($F_{1,148} = 42.0$, $P < 0.001$). For invasive range genotypes, plants
213 that flowered at intermediate times produced the most seeds. However, for native range
214 genotypes, the plants that flowered the earliest produced the most seeds (Fig. 4C).

215

216 **Discussion**

217 We found differences in traits between genotypes collected from the native and
218 invasive ranges of *Medicago polymorpha*. That all plants were grown in a common
219 environment suggests that these traits are the result of genetic differences among native and

220 invasive range plants that have evolved during or after the invasion process. Overall, invasive
221 range genotypes tended to flower sooner, produced more fruits and seeds, and produced less
222 biomass, primarily because of relatively less investment in aboveground growth relative to
223 native genotypes. Such evolution could have occurred because of genetic drift if the
224 introduction and success of individual genotypes was random. However, consistent
225 differences between invasive and native genotypes suggest that selection was more likely to
226 have driven evolutionary changes. We observed differences among ranges, but also
227 considerable variation within ranges, suggesting that both drift and selection may have played
228 a role in evolution of these traits during invasion.

229 Invasive genotypes tended to have higher fecundity than native genotypes, producing
230 greater numbers of fruits and seeds. Increased fecundity is often associated with increased
231 invasion success because greater propagule pressure increases the likelihood of success by
232 some of those propagules (Simberloff, 2009; Lockwood et al., 2009). Jelbert et al. (2015)
233 found that invasive plant species tended to be larger and had higher fecundity than closely
234 related non-invasive plants, which has been demonstrated further in both invasive plant and
235 animal species (Rejmánek, 2020). However, other studies have found exceptions to this rule.
236 Although invasive genotypes of *Hypericum perforatum* quickly adapted to new
237 environmental conditions, there was no difference in fecundity or size between invasive and
238 native genotypes (Maron et al., 2004). Rather, in some cases, phenotypic plasticity, rather
239 than fecundity, may be the key trait driving invasion success (Turner et al., 2015; Nelson &
240 Anderson, 2015). The fitness advantages of producing more seeds are often limited if higher
241 quality seeds have a greater chance of germination or survival (Wen & Simons, 2020).
242 Although we did not observe such a trade-off between seed weight and seed number, we did
243 observe more variation in seed weight than in seed number and the lack of trade-off may
244 reflect separate processes influencing the evolution of these two traits.

245 We found marginally significant differences in flowering time, biomass, and
246 above:belowground biomass ratio between invasive and native ranges. The statistics suggest
247 there is a >90-95% chance that differences in our sample represent actual differences between
248 populations, so here we interpret these as real differences, but also note that these results
249 should be interpreted with caution because of significant variation within invasive and native
250 ranges. Invasive genotypes flowered earlier than native genotypes, which may provide them
251 with a reproductive advantage in highly seasonal environments where plants must reproduce
252 quickly before conditions become unfavorable (Badri et al., 2016; Helliwell et al., 2018). We
253 also found that genotypes collected from the invasive range tended to be smaller than
254 genotypes collected from the native range, largely due to relatively less investment in
255 aboveground growth. This is contrary to many findings of increased size in the invasive range
256 (Van Kleunen et al., 2010). Increased size in invasive plants is often thought to be a result of
257 release from expensive investments in defense against natural enemies, and that energy can
258 be reinvested into growth that confers increased competitive ability (Blossey & Notzold,
259 1995). Although increased size is often used as a proxy for competitive ability, previous work
260 on this species suggests that competitive ability is not well correlated with size (Getman-
261 Pickering et al. 2018). Even if one assumed such a correlation, whether increased size confers
262 greater competitive ability against native species in the introduced range would likely depend
263 on whether competition occurs above or belowground. If competitors are indeed the agent of
264 selection on size in the invasive range of *M. polymorpha*, then the increased investment in
265 belowground biomass by invasive genotypes suggests that size is driven by competition for
266 nutrients or rhizobia in the soil, rather than aboveground competition for light or space
267 (Wang et al., 2017; Eisenhauer et al., 2017).

268 However, there may be other agents of selection on belowground growth, including
269 access to rhizobial mutualists or nutrient acquisition. The different strategies displayed by

270 invasive and native genotypes may indicate that successful *M. polymorpha* invaders dedicate
271 more resources to root growth, which may provide more nutrients to provision the highest
272 possible number of fruits and seeds. This is in contrast to successful genotypes in the native
273 range of *M. polymorpha*, which invest more resources into aboveground biomass production,
274 perhaps at the cost of producing fewer fruit and seeds. Along a continuum between r and K
275 life history strategies (MacArthur & Wilson, 1967), native range *M. polymorpha* may be
276 more like K-strategists, occupying a more stable ecosystem, and invasive genotypes may be
277 more like r-strategists, moving between more unpredictable, potentially unstable ecosystems.
278 Just as invasive species tend to fall more toward the r end of the r-K continuum, those
279 genotypes within a species that fall closer to the r end of the continuum may experience
280 positive selection during invasion. Although this weedy strategy may give each individual
281 seed a lower chance of survival, it may allow greater net fitness of genotypes that cast a wider
282 net across microclimates in novel environments.

283 In addition to the observed trait differences between invasive and native ranges of *M.*
284 *polymorpha*, there were also large differences among genotypes within ranges. This confirms
285 previous work in this species that documented different phenotypes among individuals,
286 including plant height, trichome number, coloration, and fruit morphology (Bullitta et al.,
287 1994; Graziano et al., 2009; Badri et al., 2016). In addition to the traits documented here, we
288 similarly observed differences in leaf pattern and color among genotypes that we did not
289 quantify in this study. In addition to latitudinal clines in phenotypes in both the native and a
290 portion of the invasive range, two distinct subpopulations of *M. polymorpha* have been
291 identified in the native range (Helliwell et al., 2018), perhaps explaining some intra-range
292 variation among genotypes. Additionally, there is likely to be some population structure
293 within the invasive range, as many invasive range populations are isolated by large distances,
294 but may also share the same source of introduction. Most genotypes in North and South

295 America appear to have been introduced from the western Mediterranean region (Helliwell et
296 al. 2018), but the origin of invasive genotypes on other continents is not yet known. Our
297 results do not indicate that phenotypes in the broad invasive range are a subset of native range
298 phenotypes, so it is possible that other continents have different origin sources. As most
299 invasive populations are the result of escape from agriculture, knowing the source of cultivars
300 in each location may explain some of this genotypic variation. Population structure will also
301 likely depend on the time since the start of invasion and how long populations have been
302 isolated from one another.

303 We observed non-linear relationships between fitness and three traits (biomass,
304 biomass allocation, and flowering phenology), but these relationships were stronger in
305 invasive range genotypes than in native range genotypes. For example, genotypes from the
306 invasive range that flower too early or too late have lower fecundity, but for native range
307 genotypes, only later flowering plants had lower fecundity. One must be cautious in inferring
308 selection gradients from these patterns, as these genotypes were not collected from the same
309 population and likely do not reflect variation within any single population. However, they
310 were grown in the same environment in this study, albeit an artificial one. Taken at face
311 value, these data suggest stabilizing selection in the invasive range, but either directional
312 selection or no selection on the same traits in the native range. It would be useful to know if
313 these patterns hold up in natural environments, and what, if any, the agents of stabilizing
314 selection may be in those environments, and whether those agents of selection differ between
315 native and invasive ranges.

316 These trait differences between invasive and native ranges provide evidence of which
317 traits may have allowed *M. polymorpha* to successfully invade non-native habitats.
318 Understanding the traits that allow plants to become invasive can enhance strategies to limit
319 or prevent invasion. Differences in traits like flowering phenology, fecundity, biomass, and

320 biomass allocation may have important implications for how to control invasion spread.
321 Moreover, our results suggest that understanding how these traits evolve during or after
322 invasion may be critical to understanding how plants are able to initially colonize new
323 environments, or proliferate after they have been introduced. Understanding these
324 evolutionary changes on ecological time scales in more invasive species will provide a
325 broader understanding of the role of evolution in invasion dynamics.

326

327 **Data Availability Statement**

328 The data for this study will be made available on Dryad prior to publication.

329

330 **References**

- 331 Agashe, D., & Bolnick, D. I. (2010). Intraspecific genetic variation and competition interact
332 to influence niche expansion. *Proceedings of the Royal Society B: Biological Sciences*,
333 277(1696), 2915–2924.
- 334 Badri, M., Cheikh, N. Ben, Mahjoub, A., & Abdelly, C. (2016). Morpho-phenological
335 diversity among natural populations of *Medicago polymorpha* of different Tunisian
336 ecological areas. *African Journal of Biotechnology*, 15(25), 1330–1338.
- 337 Barrett, S. C. H., Colautti, R. I., & Eckert, C. G. (2008). Plant reproductive systems and
338 evolution during biological invasion. *Molecular Ecology*, 17(1), 373–383.
- 339 Bayliss, S. L. J., terHorst, C. P., & Lau, J. A. (2017). Testing genotypic variation of an
340 invasive plant species in response to soil disturbance and herbivory. *Oecologia*, 183(4),
341 1135–1141.
- 342 Blossey, B., & Notzold, R. (1995). Evolution of Increased Competitive Ability in Invasive
343 Nonindigenous Plants: A Hypothesis. *The Journal of Ecology*, 83(5), 887.
- 344 Bolnick, D. I., Amarasekare, P., Araújo, M. S., Bürger, R., Levine, J. M., Novak, M., Rudolf,

345 V. H. W., Schreiber, S. J., Urban, M. C., & Vasseur, D. A. (2011). Why intraspecific
346 trait variation matters in community ecology. *Trends in Ecology and Evolution*, 26(4),
347 183–192.

348 Bolnick, D. I., Svanbäck, R., Fordyce, J. A., Yang, L. H., Davis, J. M., Hulsey, C. D., &
349 Forister, M. L. (2003). The ecology of individuals: incidence and implications of
350 individual specialization. *The American Naturalist*, 161(1), 1–28.

351 Bullitta, S., Floris, R., Hayward, M. D., Loi, A., Porqueddu, C., & Veronesi, F. (1994).
352 *Morphological and biochemical variation in Sardinian populations of Medicago*
353 *polymorpha L. suitable for rainfed mediterranean conditions* (pp. 223–232). Springer,
354 Dordrecht.

355 Davidson, A. M., Jennions, M., & Nicotra, A. B. (2011). Do invasive species show higher
356 phenotypic plasticity than native species and, if so, is it adaptive? A meta-analysis.
357 *Ecology Letters*, 14(4), 419–431.

358 Eisenhauer, N., Lanoue, A., Strecker, T., Scheu, S., Steinauer, K., Thakur, M. P., &
359 Mommer, L. (2017). Root biomass and exudates link plant diversity with soil bacterial
360 and fungal biomass. *Scientific Reports*, 7(1), 1–8.

361 Geng, Y., van Klinken, R. D., Sosa, A., Li, B., Chen, J., & Xu, C. Y. (2016). The relative
362 importance of genetic diversity and phenotypic plasticity in determining invasion
363 success of a clonal weed in the USA and China. *Frontiers in Plant Science*,
364 7(FEB2016), 213.

365 Getman-Pickering, Z. L., terHorst, C. P., Magnoli, S. M., & Lau, J. A. (2018). Evolution of
366 increased *Medicago polymorpha* size during invasion does not result in increased
367 competitive ability. *Oecologia*, 188(1), 203–212.

368 Graziano, D., Giorgio, G. Di, Ruisi, P., Amato, G., Giambalvo, D., Graziano, D., Giorgio, G.
369 Di, Ruisi, P., Amato, G., & Giambalvo, D. (2009). Variation in pheno-morphological

370 and agronomic traits among burr medic (*Medicago polymorpha* L.) populations
371 collected in Sicily, Italy. *Crop and Pasture Science*, 61(1), 59–69.

372 Hairston, N. G., Ellner, S. P., Geber, M. A., Yoshida, T., & Fox, J. A. (2005). Rapid
373 evolution and the convergence of ecological and evolutionary time. *Ecology Letters*,
374 8(10), 1114–1127.

375 Helliwell, E. E., Faber-Hammond, J., Lopez, Z. C., Garoutte, A., von Wettberg, E., Friesen,
376 M. L., & Porter, S. S. (2018). Rapid establishment of a flowering cline in *Medicago*
377 *polymorpha* after invasion of North America. *Molecular Ecology*, 27(23), 4758–4774.

378 Hughes, A. R., Inouye, B. D., Johnson, M. T. J., Underwood, N., & Vellend, M. (2008).
379 Ecological consequences of genetic diversity. *Ecology Letters*, 11(6), 609–623.

380 Hui, C., Richardson, D. M., Landi, P., Minoarivelo, H. O., Garnas, J., & Roy, H. E. (2016).
381 Defining invasiveness and invasibility in ecological networks. *Biological Invasions*,
382 18(4), 971–983.

383 Jelbert, K., Stott, I., McDonald, R. A., & Hodgson, D. (2015). Invasiveness of plants is
384 predicted by size and fecundity in the native range. *Ecology and Evolution*, 5(10), 1933.

385 Keane, R. M., & Crawley, M. J. (2002). Exotic plant invasions and the enemy release
386 hypothesis. *Trends in Ecology & Evolution*, 17(4), 164–170.

387 Lambrinos, J. G. (2004). How interactions between ecology and evolution influence
388 contemporary invasion dynamics. *Ecology*, 85(8), 2061–2070.

389 Lau, J. A., & Strauss, S. Y. (2005). Insect herbivores drive important indirect effects of exotic
390 plants on native communities. *Ecology*, 86(11), 2990–2997.

391 Lavergne, S., & Molofsky, J. (2007). Increased genetic variation and evolutionary potential
392 drive the success of an invasive grass. *Proceedings of the National Academy of Sciences*,
393 104(10), 3883–3888.

394 Lockwood, J. L., Cassey, P., & Blackburn, T. M. (2009). The more you introduce the more

395 you get: the role of colonization pressure and propagule pressure in invasion ecology.
396 *Diversity and Distributions*, 15(5), 904–910.

397 MacArthur, R. H., & Wilson, E. O. (1967). The Theory of Island Biogeography. In *Nature*
398 (Vol. 413, Issue 6857).

399 Marchini, G. L., Maraist, C. A., & Cruzan, M. B. (2019). Trait divergence, not plasticity,
400 determines the success of a newly invasive plant. *Annals of Botany*, 123(4), 667–679.

401 Maron, J. L., Klironomos, J., Waller, L., & Callaway, R. M. (2014). Invasive plants escape
402 from suppressive soil biota at regional scales. *Journal of Ecology*, 102(1), 19–27.

403 Maron, J. L., Vilà, M., Bommarco, R., Elmendorf, S., & Beardsley, P. (2004). Rapid
404 evolution of an invasive plant. *Ecological Monographs*, 74(2), 261–280.

405 Nelson, M. F., & Anderson, N. O. (2015). Variation Among Genotypes and Source Habitats
406 in Growth and Fecundity of the Wetland Invasive Plant *Phalaris arundinacea* L.
407 *Wetlands 2015 35:6*, 35(6), 1175–1184.

408 Palacio-López, K., & Gianoli, E. (2011). Invasive plants do not display greater phenotypic
409 plasticity than their native or non-invasive counterparts: a meta-analysis. *Oikos*, 120(9),
410 1393–1401.

411 Parker, I. M., & Gilbert, G. S. (2007). When there is no escape: The effects of natural
412 enemies on native, invasive, and noninvasive plants. *Ecology*, 88(5), 1210–1224.

413 Porter, S. S., & Simms, E. L. (2014). Selection for cheating across disparate environments in
414 the legume-rhizobium mutualism. *Ecology Letters*, 17(9), 1121–1129.

415 Rejmánek, M. (2020). Invasiveness. *Encyclopedia of Biological Invasions*, 379–385.

416 Schoener, T. W. (2011). The Newest Synthesis: Understanding the Interplay of Evolutionary
417 and Ecological Dynamics. *Science*, 331(6016), 426–429.

418 Simberloff, D. (2009). The Role of Propagule Pressure in Biological Invasions.
419 [Http://Dx.Doi.Org/10.1146/Annurev.Ecolsys.110308.120304](http://dx.doi.org/10.1146/annurev.ecolsys.110308.120304), 40, 81–102.

420 terHorst, C. P., & Lau, J. A. (2015). Genetic variation in invasive species response to direct
421 and indirect species interactions. *Biological Invasions*, 17(2), 651–659.

422 terHorst, C. P., Wirth, C., & Lau, J. A. (2018). Genetic variation in mutualistic and
423 antagonistic interactions in an invasive legume. *Oecologia*, 188(1), 159–171.

424 terHorst, C. P., Zee, P. C., Heath, K. D., Miller, T. E., Pastore, A. I., Patel, S., Schreiber, S. J.,
425 Wade, M. J., & Walsh, M. R. (2018). Evolution in a Community Context: Trait
426 Responses to Multiple Species Interactions*. <https://doi.org/10.1086/695835>, 191(3),
427 368–380.

428 Thompson, J. N. (1998). Rapid evolution as an ecological process. *Trends in Ecology &*
429 *Evolution*, 13(8), 329–332.

430 Turner, K. G., Fréville, H., & Rieseberg, L. H. (2015). Adaptive plasticity and niche
431 expansion in an invasive thistle. *Ecology and Evolution*, 5(15), 3183–3197.

432 Van Kleunen, M., Weber, E., & Fischer, M. (2010). A meta-analysis of trait differences
433 between invasive and non-invasive plant species. *Ecology Letters*, 13(2), 235–245.

434 Wang, X. Y., Ge, Y., & Wang, J. (2017). Positive effects of plant diversity on soil microbial
435 biomass and activity are associated with more root biomass production. *Journal of Plant*
436 *Interactions*, 12(1), 533–541.

437 Wen, L., & Simons, A. M. (2020). Delusions of grandeur: Seed count is not a good fitness
438 proxy under individual variation in phenology. *Journal of Evolutionary Biology*, 33(8),
439 1039–1049.

440 Westbrooks, R. (1998). Jointed Goatgrass Invasive plants: Changing the landscape of
441 America. *All U.S. Government Documents (Utah Regional Depository)*, 22-23 ST-
442 Jointed Goatgrass Invasive plants: Cha.

443 Zenni, R. D., & Nuñez, M. A. (2013). The elephant in the room: The role of failed invasions
444 in understanding invasion biology. *Oikos*, 122(6), 801–815.

445

446 **Statements and Declarations**

447 We thank E. Sharma for help in the greenhouse during this experiment and R. Mackelprang,
448 N. Silbiger, and J. Yoder for valuable input on previous versions of this manuscript. This
449 work was supported by grants from California State University, Northridge to Carmen
450 Hoffbeck and from the National Science Foundation to Casey terHorst (OCE-1559105 and
451 DEB-1754449).

452 The authors have no relevant financial or non-financial interests to disclose.

453 All authors contributed to the experimental concept and design. Preparation, data collection,
454 and data analysis were performed by Carmen Hoffbeck. The first manuscript draft was
455 written by Carmen Hoffbeck. All authors edited, read, and approved the final manuscript.

Figures

Figure 1

(A) Mean days to first flower (\pm SEM) of invasive and native range genotypes and (B) variation among genotypes within each range.

Figure 2

(A) Mean total biomass (\pm SEM) of invasive and native range *M. polymorpha* genotypes and (B) variation among genotypes within each range. (C) Mean above:belowground biomass ratios (\pm SEM) of invasive and native range genotypes and (D) variation among genotypes within each range.

Figure 3

(A) Mean seed number (\pm SEM) of *M. polymorpha* invasive and native range genotypes and (B) variation among genotypes within each range.

Figure 4

Non-linear relationships between *M. polymorpha* seed number and (A) total biomass, (B) above:belowground biomass ratio, and (C) days to flower, for invasive and native range genotypes.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [SupplementalMaterial.pdf](#)