Received: 28 June 2022 | Accepted: 24 July 2022
DOI: 10.1111/1365-2745.13981

Journal of Ecology BRITISH ECOLOGICAL SOCIETY

RESEARCH ARTICLE

Spatiotemporal dynamics of genetic variation at the quantitative and molecular levels within a natural *Arabidopsis thaliana* population

Belén Méndez-Vigo[1] | Antonio R. Castilla[2] | Rocío Gómez[3] | Arnald Marcer[4,5] | Carlos Alonso-Blanco[1] | F. Xavier Picó[3]

[1]Departamento de Genética Molecular de Plantas, Centro Nacional de Biotecnología (CNB), Consejo Superior de Investigaciones Científicas (CSIC), Madrid, Spain

[2]Department of Biology, McCoy College of Science, Mathematics & Engineering, Midwestern State University, Wichita Falls, Texas, USA

[3]Departamento de Ecología Integrativa, Estación Biológica de Doñana (EBD), Consejo Superior de Investigaciones Científicas (CSIC), Sevilla, Spain

[4]CREAF, E08193 Bellaterra (Cerdanyola del Vallès), Catalonia, Spain

[5]Universitat Autònoma de Barcelona, E08193 Bellaterra (Cerdanyola del Vallès), Catalonia, Spain

Correspondence
F. Xavier Picó
Email: xpico@ebd.csic.es

Funding information
Agencia Estatal de Investigación (AEI), Grant/Award Number: CGL2016-77720-P and PID2019-104135GB-I00

Handling Editor: Richard Shefferson

[Correction added on 31-Dec-2022, after first online publication: The copyright line was changed.]

Abstract

1. Evolutionary change begins at the population scale. Therefore, understanding adaptive variation requires the identification of the factors maintaining and shaping standing genetic variation at the within-population level. Spatial and temporal environmental heterogeneity represent ecological drivers of within-population genetic variation, determining the evolutionary trajectory of populations along with random processes. Here, we focused on the effects of spatiotemporal heterogeneity on quantitative and molecular variation in a natural population of the annual plant *Arabidopsis thaliana*.
2. We sampled 1093 individuals from a Spanish *A. thaliana* population across an area of 7.4 ha for 10 years (2012–2021). Based on a sample of 279 maternal lines, we estimated spatiotemporal variation in life-history traits and fitness from a common garden experiment. We genotyped 884 individuals with nuclear microsatellites to estimate spatiotemporal variation in genetic diversity. We assessed spatial patterns by estimating spatial autocorrelation of traits and fine-scale genetic structure. We analysed the relationships between phenotypic variation, geographical location and genetic relatedness, as well as the effects of environmental suitability and genetic rarity on phenotypic variation.
3. The common garden experiment indicated that there was more temporal than spatial variation in life-history traits and fitness. Despite the differences among years, genetic distance in ecologically relevant traits (e.g. flowering time) tended to be positively correlated to genetic distance among maternal lines, while isolation by distance was less important. Genetic diversity exhibited significant spatial structure at short distances, which were consistent among years. Finally, genetic rarity, and not environmental suitability, accounted for genetic variation in life-history traits.
4. *Synthesis*. Our study highlighted the importance of repeated sampling to detect the large amount of genetic diversity at the quantitative and molecular levels

---

Belén Méndez-Vigo and Antonio R. Castilla contributed equally to this work.

that a single *A. thaliana* population can harbour. Overall, population genetic attributes estimated from our long-term monitoring scheme (genetic relatedness and genetic rarity), rather than biological (dispersal) or ecological (vegetation types and environmental suitability) factors, emerged as the most important drivers of within-population structure of phenotypic variation in *A. thaliana*.

KEYWORDS
Arabidopsis thaliana, environmental suitability, fitness, genetic rarity, genetic relatedness, life-history traits, spatial structure, within-population variation

1 | INTRODUCTION

The spatiotemporal dynamics of genetic variation affects the evolutionary trajectory of plant populations. This is because genetic variation defines the spectrum of phenotypic variation for selection to act upon. Furthermore, adaptive evolution is accepted to be mostly determined by standing genetic variation, rather than by new mutations, as selection acts faster on available variation starting at higher frequencies (Barrett & Schluter, 2008; Fournier-Level et al., 2011; but see Fulgione et al., 2022). Hence, understanding how natural populations maintain genetic variation has been a major goal in evolution (Barrett & Schluter, 2008; Barton & Keightley, 2002; Friedman et al., 2019; Gillespie & Turelli, 1989; Mitchell-Olds et al., 2007; Mojica et al., 2012; Wittmann et al., 2017). At present, heterogeneous selection, along with epistatic interactions among loci, is believed to account for the abundant genetic variation commonly found in populations, which cannot be explained solely by mutation–selection balance (Mitchell-Olds et al., 2007; Weinig et al., 2003).

From an ecological viewpoint, there are two major sources of environmental heterogeneity underlying within-population genetic variation. On the one hand, the diversity of physical and biological environments in a population may promote spatially heterogeneous selection, leading to fine-scale genetic differentiation in fitness-related traits (Argyres & Schmitt, 1991; Galloway, 1995; Kalisz, 1986; Mitchell-Olds & Bergelson, 1990; Paccard et al., 2013; Prati & Schmid, 2000; Schemske, 1984; Stratton & Bennington, 1996). Fine-scale genetic structure is commonly detected at the molecular level in various plant species (Castilla et al., 2019; Linhart & Grant, 1996; Vekemans & Hardy, 2004; Volis et al., 2016). This is because spatial genetic differentiation due to divergent selection can be enhanced when acting in concert with limited gene flow and selection operating on multiple traits (Barton & Turelli, 1989). Finally, the implications of small-scale biotic and abiotic heterogeneity may scale up to the entire community by increasing species diversity, as spatial heterogeneity has the potential to buffer populations and communities from environmental alterations (De Frenne et al., 2013; Denney et al., 2020; McLaughlin et al., 2017).

On the other hand, temporal heterogeneity refers to the ubiquitous year-to-year variation in environmental conditions affecting the window of opportunity for completing the life cycle, particularly in annuals. Long-term monitoring of natural plant populations (Ehrlén & Valdés, 2020; Quintana-Ascencio et al., 2018) represents a powerful means to assess the ecological and evolutionary consequences of temporal heterogeneity. On top of that, experimental approaches allow the quantification of temporal change in genetic variation by comparing past and contemporary performance of plant populations at different points in time encompassing several generations (i.e. resurrection experiments; Franks et al., 2007; Nevo et al., 2012; Kuester et al., 2016, Gómez et al., 2018). Overall, long-term monitoring schemes and resurrection experiments reveal how fast plant populations can change genetically over time. In addition, empirical evidence supports the accepted paradigm by which temporal heterogeneity in environmental conditions may result in varying selection (Ågren et al., 2017; Exposito-Alonso et al., 2018; Fournier-Level et al., 2013; Siepielski et al., 2009; Wittmann et al., 2017). In particular, temporal heterogeneity promotes the maintenance of genetic variation in populations by affecting the adaptive response of genotypes to selection differences over time. Overall, both spatial and temporal heterogeneity may also have indirect demographic effects on plant populations, as they create subpopulations reducing effective population size and determining the extent of stochastic processes shaping genetic variation.

In this study, we tackled both sources of environmental heterogeneity simultaneously to disentangle their role in structuring genetic variation at the quantitative and molecular levels within a plant population. To this end, we quantified the spatiotemporal environmental heterogeneity in a natural population of the annual *Arabidopsis thaliana* and its effects on variation in life-history traits and genetic diversity. In particular, we sampled a single natural *A. thaliana* population in central Spain across an area of 7.4 ha for 10 years (2012–2021). Over this period, we collected seeds from 1093 geo-referenced individuals across the population. Occurrence data were used to compute an environmental suitability index at the within-population scale. If the intensity of selection increases in unsuitable areas, we would expect a relationship between variation in environmental suitability and variation in life-history traits with a higher effect on fitness. We estimated spatiotemporal variation in life-history traits and fitness from a common garden experiment

(279 maternal lines; 2012–2015) and molecular variation with nuclear microsatellites (884 individuals; 2012–2020). Molecular data were used to compute a genetic rarity index at the within-population scale, pinpointing areas with combinations of low or common frequency alleles. If genetic rarity relates to the occurrence of novel genetic variants (e.g. mutants, migrants and their descendants) with a higher degree of adaptation or maladaptation, we would expect a relationship between variation in genetic rarity and variation in life-history traits under selection.

Previous studies showed that *A. thaliana* exhibits substantial within-population variation in quantitative traits (Brachi et al., 2013; Frachon et al., 2017; Gómez et al., 2018; Kuittinen et al., 1997; Méndez-Vigo et al., 2013; Stenøien et al., 2005) and molecular markers (Bomblies et al., 2010; Castilla et al., 2020; Falahati-Anbaran et al., 2014; Gomaa et al., 2011; Gómez et al., 2018; Le Corre, 2005). In contrast, our understanding of the spatiotemporal dynamics of genetic variation within natural populations is scarce. In fact, the bulk of evolutionary research on *A. thaliana* has mostly used genetic constructs (e.g. recombinant inbred lines; El-Lithy et al., 2004; Bentsink et al., 2010; Sánchez-Bermejo et al., 2012; Ågren et al., 2013, 2017; Postma & Ågren, 2016; Taylor et al., 2017) and accession collections (Atwell et al., 2010; Brachi et al., 2010; Fournier-Level et al., 2011; Hancock et al., 2011; Wilczek et al., 2014; The 1001 Genomes Consortium, 2016; Tabas-Madrid et al., 2018; Exposito-Alonso et al., 2019; Martínez-Berdeja et al., 2020; Fulgione et al., 2022). Both approaches estimate the effect of large-scale environmental differences on phenotypic variation, pinpoint the major genes influencing trait variation and disentangle the genetic basis of quantitative traits underlying local adaptation. In contrast, they cannot account for the amount, distribution and maintenance of genetic variation within populations.

Here, we addressed the following specific questions. First, what is the extent of within-population spatial and temporal variation in life-history traits and molecular markers in *A. thaliana*? We know that *A. thaliana* populations exhibit temporal genetic variation in fitness-related traits, such as flowering time, and in their genetic composition (Frachon et al., 2017; Gómez et al., 2018). In contrast, our comprehension of the effects of within-population spatial environmental heterogeneity on genetic variation is scarce (but see Bell & Lechowicz, 1991; Stratton & Bennington, 1996; Frachon et al., 2017). Second, what is the role of genetic relatedness among individuals in structuring variation in life-history traits and fitness within an *A. thaliana* population? Given the limited *A. thaliana*'s dispersal ability and its self-fertilizing habit (Bomblies et al., 2010; Castilla et al., 2020; Falahati-Anbaran et al., 2014; Lundemo et al., 2009), we expect a strong spatial structure of within-population phenotypic variation mediated by dispersal limitations and the subsequent increase in genetic relatedness among neighbouring *A. thaliana* individuals. And third, can environmental suitability and genetic rarity, two parameters estimated from the long-term monitoring scheme, provide insight into spatiotemporal variation in life-history traits and fitness in an *A. thaliana* population? We ignore whether occurring in cold spot or hot spot for environmental suitability and genetic rarity may account for within-population phenotypic variation in *A. thaliana*.

2 | MATERIALS AND METHODS

2.1 | Study population and environmental data

The Moral de Calatrava population is located in central Spain (Ciudad Real province; 38.8743°N, 3.5296°W; altitude: 719–729 m; Figure 1a). Climate is continental Mediterranean with hot dry summers, cold winters and a marked seasonal precipitation. The population has an annual mean temperature of 14.3°C, with a minimum and maximum mean temperature of 0.1°C in January and of 34.0°C in July, respectively. On average, annual total precipitation is of 414.7 mm, with a minimum and maximum values of 10.3 mm in August and of 51.0 mm in December, respectively (Digital Climatic Atlas from the Iberian Peninsula; data: 1950–1999; accessed on February 9, 2020).

The hilly landscape around the *A. thaliana* population is dominated by vineyards, olive groves and cereal fields, and scattered by patches of natural vegetation. The study population is located in one of these patches (Figure 1b). We did not detect any substantial change in the landscape during this study (2012–2021). In fact, the site has remained undisturbed by wildfires or land use changes at least since 1997 (Castilla et al., 2020). We only detected small-scale disturbances caused by rabbits, *Oryctolagus cuniculus*, which construct burrows for shelter and nesting, and wild boars, *Sus scrofa* that dig up the soil for food.

There are different vegetation types at the *A. thaliana* population. Scrubland is mainly composed of dominant Mediterranean shrub species (Figure 1c), such as rockroses of the genus *Cistus* (e.g. *C. ladanifer* L. and *C. monspeliensis* L.) and the rosemary, *Rosmarinus officinalis* L. Holm oak, *Quercus ilex* L. stands occur across the site, creating shadier environments than the rest of vegetation types (Figure 1d). Torrent dry riverbeds cross over the population receiving runoff water after rains. As a result, dry riverbeds are stony with no or scarce vegetation (Figure 1e). The grassland is an open space with a different plant community, composed of forbs, herbs and grasses.

We obtained meteorological data for the study period (2012–2021) from the Agencia Estatal de Meteorología of Spain. Data included daily records of minimum and maximum temperatures and precipitation averaged from the four nearest automatic meteorological stations to the *A. thaliana* population (Figure S1; mean ± SE distance to the population = 18.7 ± 2.6 km; range = 12–29 km). These automatic meteorological stations were located at a similar altitude (range = 622–731 m a.s.l.). We also recorded daily minimum and maximum temperature with HOBO Pendant UA-002-08 temperature loggers (Onset Computer Corporation, Inc.) between July 2018 and October 2020 (Figure S2). We placed six data loggers across the population to quantify temperature differences among main vegetation types (dry riverbed, scrubland and holm oak), which were determined by the proportion of each vegetation type in a circular

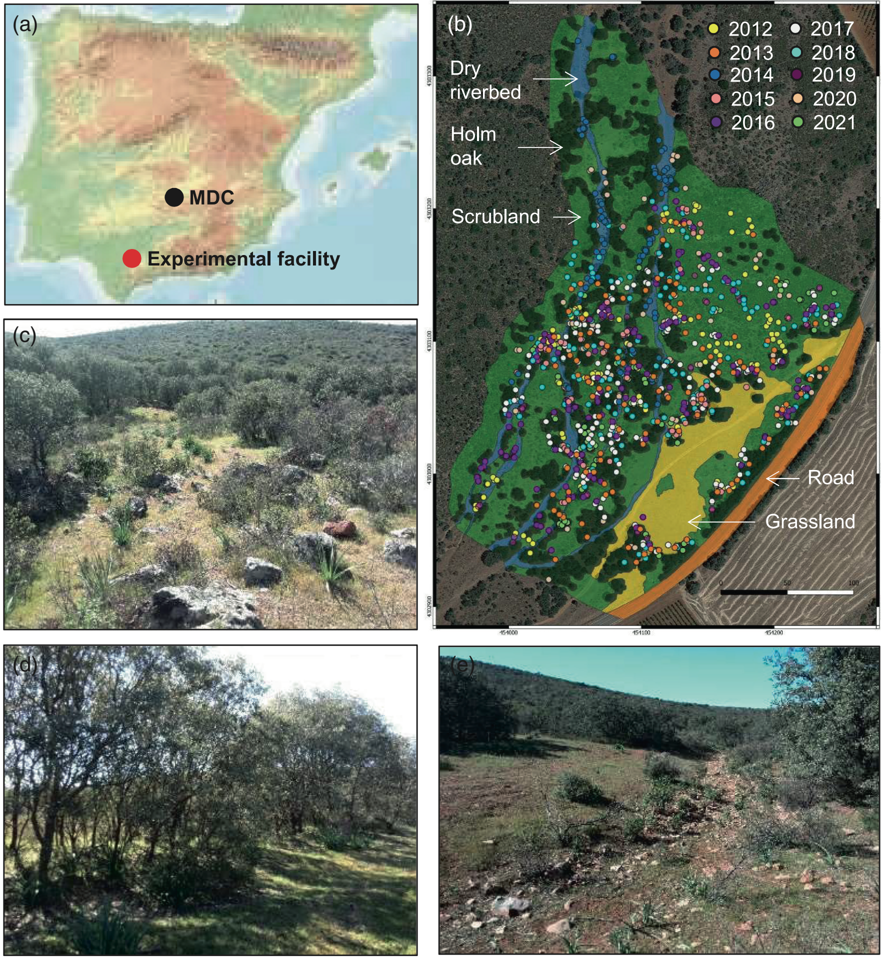

FIGURE 1 Vegetation types and distribution of *Arabidopsis thaliana* individuals across the Moral de Calatrava population. (a) Location of the *A. thaliana* population and the experimental facility in the Iberian Peninsula. (b) Vegetation types digitized onto an orthophotograph of the *A. thaliana* population with the distribution of sampled individuals per year. Scale: 0–100 m. (c) Photograph of the scrubland. (d) Photograph of a holm oak stand. (e) Photograph of a dry riverbed.

area (5 m radius) around its global positioning system (GPS) coordinate. We suspended data loggers on rockrose or holm oak branches 20–30 cm above-ground. We did not use any shielding because we deployed sensors in natural shade.

2.2 | Field sampling

During 10 years (2012–2021), we sampled the *A. thaliana* population across its 7.42 ha in the first half of May. Every year, we carried out the field sampling as follows. We divided the population into sections using landmarks (Figure S3) and a few GPS coordinates at the north and south population margins. The west and east margins were clearly delimited by a pronounced change in slope and by the road, respectively. Within each section, at least two persons searched for *A. thaliana* individuals following a non-fixed zigzag path until covering the entire section before moving to the next section. Completing the sampling across the entire population required up to a full day (8–9 h) in years when *A. thaliana* was abundant. We used a GPS (Garmin International, Inc.; positional error: 4 m) to record the longitude and latitude of sampled individuals. Whenever possible, every year we collected seeds from individuals separated from each other by 5–10 m (mean ± SE distance between individual pairs = 116.9 ± 1.8 m; range = 1.0–406.6 m). Overall, we sampled 1093 *A. thaliana* individuals (Figure 1b), ranging from a low of 54 in 2019 to a high of 164 in 2017. We did not discriminate among vegetation types during the sampling because vegetation types were assigned after the sampling (see below). Thus, the final number of individuals per year reflected the temporal variation in *A. thaliana* occurrence among vegetation types and represented an indirect estimate of *A. thaliana* abundance. In years when *A. thaliana* was not abundant, sample size was low, whereas in years when the species was almost ubiquitous, subsequent sample sizes were higher. Our estimates of plant abundance in good years (e.g. 2017), counting all reproductive individuals found within ca. 80 plots (5 m radius) haphazardly set up across the population, yielded an estimated population size above 10,000 individuals (F.X. Picó, unpublished data). In poor years, above-ground population size could be in the range of just a few hundred (F.X. Picó and C. Alonso-Blanco, pers. obs.). After each field campaign, we multiplied field-collected seeds by the single seed descent method in a glasshouse at the Centro Nacional de Biotecnología (Madrid, Spain). Multiplied seeds were stored in cellophane bags, in dry conditions, at room temperature and in darkness. In these conditions, *A. thaliana* seeds remain viable for years (F.X. Picó and C. Alonso-Blanco, pers. comm.).

2.3 | Vegetation types

We quantified the percentage of vegetation types at the *A. thaliana* population by digitizing an aerial orthophotograph from 2015 (Portal de Mapas de la Junta de Castilla-La Mancha; accessed on 25 June 2018) with QGIS v.3.4 (https://www.qgis.org/en/site/). We categorized 48.38% of the area as scrubland, 30.63% as holm oak, 8.95% as grassland, 7.64% as dry riverbed and 4.41% as road (Figure 1b). We used the geographical coordinates to assign a vegetation type to each sampled individual from the digitized orthophotograph. We calculated the proportion of each vegetation type in a circular area (1 m radius) around the GPS coordinate of each individual. We assigned individuals to their vegetation type based on the highest membership proportion. Such assignment was straightforward because individual membership percentages were high: mean (±SE) values were of 95.0% (±0.5) for scrubland (*N* = 553 individuals), 90.9% (±0. 9) for holm oak (*N* = 335), 89.5% (±1.1) for dry riverbed (*N* = 179) and 88.1% (±3.7) for grassland (*N* = 26). In October 2020, we validated vegetation type assignments in situ for several individuals per vegetation type, confirming that the method was accurate.

2.4 | Common garden experiment and spatiotemporal variation in phenotypic traits

We estimated variation in life-history traits and fitness from *A. thaliana* maternal lines collected during the first four sampling years (2012–2015) by undertaking a common garden experiment (Figure S4). Prior to the experiment, we obtained fresh seed with another single seed descent experiment in the greenhouse to avoid any aging or storage effects. Overall, we produced seeds from 80, 78, 76 and 45 maternal lines from 2012, 2013, 2014 and 2015, respectively, totalling 279 *A. thaliana* maternal lines. In summer 2016, we prepared nine batches of 60 seeds each per maternal line and stored them in 1.5 ml plastic tubes in darkness. We weighted three batches (279 maternal lines ×3 batches/maternal line ×60 seeds/batch = 50,220 seeds) to the nearest 0.1 mg with a Sartorious BP61S balance (Sartorius AG, Göttingen, DE).

In autumn 2016, we established the common garden experiment at the El Castillejo Botanical Garden of Sierra de Grazalema Natural Park (Cádiz province; 36.46°N, 5.30°W; altitude: 329 m; Figure S4). The experimental facility is located 291 km south of the *A. thaliana* population (Figure 1a) and represents an appropriate natural setting to estimate variation in life-history traits and fitness in *A. thaliana* from very different Iberian environments (Exposito-Alonso et al., 2018; Gómez et al., 2018; Manzano-Piedras et al., 2014; Méndez-Vigo et al., 2013). The environmental conditions at El Castillejo Botanical Garden allow plants to grow in an environment without stressful conditions, offering the opportunity to estimate the genetic component of phenotypic variation in a natural setting over the entire life cycle. Besides, previous experiments in this facility indicated that its winter conditions, albeit mild, allow vernalization of late flowering plants, even those with an obligate vernalization requirement to flower (Manzano-Piedras et al., 2014; Méndez-Vigo et al., 2013), indicating that early and late flowering *A. thaliana* individuals experience the conditions required to complete the life cycle. On average, environmental conditions at the experimental facility were warmer (annual mean temperature of 17.4°C) and wetter (total annual precipitation of 886.5 mm) than at the *A. thaliana* population of study. We sowed (November 3, 2016) six batches per maternal line in square plastic pots ($12 \times 12 \times 12\,\text{cm}^3$) filled with standard soil mixture (Abonos Naturales Cejudo Baena S.L., Utrera, ES) placed in randomized blocks. Each block included one replicate per maternal line (279 maternal lines ×6 replicates/maternal line ×60 seeds/replicate = 100,440 seeds).

We monitored the experiment weekly from the sowing day. We estimated recruitment as the maximum proportion of seedlings, obtained by dividing the maximum number of seedlings recorded per pot by 60. Recruitment occurred right after sowing. We observed the maximum recruitment within the first two surveys after seed sowing. At this facility, recruitment in *A. thaliana* is negligible after reaching the maximum number of seedlings per pot (Exposito-Alonso et al., 2018). We estimated flowering time as the number of days between sowing and flowering dates. Flowering date was given at the pot level when the majority of the plants in the pot (full-sibs with homogeneous behaviour) had the first flower open. We recorded the number of fruiting plants per pot and counted the number of fruits per plant. We estimated fecundity as the total number of seeds per plant with a function relating the number of fruits per plant with the number of seeds per fruit (Exposito-Alonso et al., 2018). We estimated survivorship as the proportion of reproductive plants relative to the maximum number of seedlings recorded. Finally, we estimated the integrated lifetime fitness (i.e. survivorship × fecundity), providing the mean number of expected seeds per plant. Losses were low in this experiment, as only 713 of 64,928 fruits (1.10%) were lost due to abortion or depredation. Diseases (unknown causes) were also low, with only 21 of 20,338 adult plants (0.10%) affected.

For each year, we computed the Moran's *I* autocorrelation coefficient to assess spatial relationships of all traits (seed weight, recruitment, flowering time, survival, fecundity and fitness). We used 60 spatial intervals with a constant number of pairwise comparisons and estimated significances from 1000 permutations with the software PASSaGE v.2 (Rosenberg & Anderson, 2011). We also estimated the correlation between pairs of traits using the Dutilleul's *t*-test, which corrects the variance and the degrees of freedom based on the extent of spatial autocorrelation, with the software SAM v.4.0 (Rangel et al., 2010).

We tested the effect of vegetation type (fixed factor), year (random factor) and maternal line (random factor) nested within year on variation in life-history traits and fitness. We excluded grassland from this analysis because only eight individuals occurred in this vegetation type (Table S1). We fitted linear mixed models (LMMs) for seed weight, flowering time, fecundity and fitness, and generalized LMMs (GLMMs) for recruitment and survival (using the Poisson distribution as the inverse link function) with the R package lme4 (Bates et al., 2015). We inspected the frequency distribution of variables, the variances of data, the existence of outliers and the model residuals to check that the major assumptions of the analyses were acceptable. We tested significances with likelihood-ratio tests. We obtained marginal (for fixed effects only) and conditional (for all effects) R^2 values for each factor and variable using the 'r2_nakagawa' function (Nakagawa et al., 2017).

To quantify the proportion of phenotypic variance accounted for by genotypic variance for all life-history traits in *A. thaliana* maternal lines collected over 4 years and grown together at the common garden experiment, we estimated broad-sense heritability (h^2) values as $h^2 = V_G/(V_G+V_E)$, where V_G is the among-maternal line variance component and V_E is the residual variance (Le Corre, 2005). We estimated all variance components and their 95% confidence intervals using the 'remlVCA' and 'VCAinference' functions of the `R` package `VCA` v.1.4.3. (https://cran.r-project.org/web/packages/VCA/index.html).

Finally, to identify the effects of selection on *A. thaliana* in maternal lines collected over 4 years and grown together at the common garden experiment, we estimated linear and quadratic selection gradients (β and γ) and selection differentials (s and C) for recruitment and flowering time using a generalized additive model to characterize a fitness function with the `R` package `gsg` (Morrissey & Sakrejda, 2013). We focused on the effects of recruitment and flowering time, the two major transitions in annuals, on fitness (survival × fecundity) in the environmental conditions of the common garden experiment. We used mean values among pots per maternal line as replicates for recruitment, flowering time and fitness. This selection analysis sought to assess whether the contribution of recruitment and flowering time to fitness estimated in a common garden experiment differed among maternal lines representing the genetic variation collected at the *A. thaliana* population over four consecutive years. To this end, we conducted all selection analyses separately by year of collection. We estimated all gradients and differentials from full models including linear and quadratic effects using standardized variables by subtracting the mean and scaling the variance. We found consistent results obtained with traditional least squares-based regressions (results not shown).

2.5 | Microsatellite genotyping and spatiotemporal variation at the molecular level

We genotyped most of the individuals collected between 2012 and 2020 at the *A. thaliana* population (N = 884 individuals) with 12 putatively neutral nuclear microsatellites (see Picó et al., 2008 for plant growing conditions, DNA extraction and marker genotyping details). Microsatellites had long allelic series (range = 8–30 alleles per marker) with an average (±SD) of 18.6 (±8.3) alleles per microsatellite. We used plants (pooling DNA from six full-sibs) obtained during the first multiplication experiment after each field campaign to extract DNA from each individual. We estimated an average genotyping error of 0.044 per locus (similar to previous estimates in *A. thaliana*; Picó et al., 2008; Gómez et al., 2018), which was obtained by genotyping twice 47 individuals.

We estimated the number of non-redundant multilocus genotypes (MLGs) with the `R` package `allelematch` (Galpern et al., 2012). We used a maximum of four mismatching alleles (the m-hat parameter) to discriminate among different MLG. All individuals were assigned to MLG, except 21 that were inconclusive. Differences in the number of unique MLG (i.e. MLG represented by one individual) among vegetation types and years were tested with χ^2 tests. For each vegetation type and year, we also computed genotypic richness and the Shannon–Wiener diversity index with the `R` package `vegan` v.2.5-6 (https://CRAN.R-project.org/package=vegan). We estimated observed (H_O) and expected heterozygosity (H_S) with `GenAlEx` v.6.5 (Peakall & Smouse, 2012). We conducted Kruskal–Wallis tests to assess whether H_O and H_S differed among vegetation types and years. Furthermore, we decomposed genetic variance among years, among individuals within years and within individuals, calculating F-statistics via the Analysis of molecular variance with `GenAlEx`. We repeated the same analysis to decompose genetic variance among vegetation types, among individuals within vegetation types and within individuals. Significance of all F-statistics was estimated by performing 1000 permutations.

For each year, we estimated the fine-scale genetic structure of *A. thaliana* individuals by computing the pairwise kinship coefficient F_{ij} (Loiselle et al., 1995) among all pairs of individuals with the software `SPAGEDI` v.1.5 (Hardy & Vekemans, 2002). We performed spatial genetic analyses using 10 distance classes (25 m each). Statistical significance of mean kinship coefficient values in each distance class was obtained by random shuffling (5000 times) individual geographical locations to define the upper and lower bounds of the 95% confidence interval around the null hypothesis (i.e. F_{ij} and pairwise spatial distances were uncorrelated). We estimated SE around mean F_{ij} values by jackknifing loci (5000 repeats). We also computed the *Sp* statistic (Vekemans & Hardy, 2004), which estimates the strength of spatial genetic structure and enables comparisons among years. We found consistent results when using classes with similar number of pairwise comparisons and variable distances (results not shown). Finally, we used the software `Programita` for spatial point-pattern analysis (Wiegand & Moloney, 2004) using the random labelling as null model to detect spatial patterns in the distribution of all individuals carrying heterozygous loci.

We correlated pairwise phenotypic distance matrices with geographical and genetic distance matrices using Mantel tests with `SAM`. Significances were estimated from 1000 permutations. We obtained the geographical distance matrix as a matrix of Euclidian distances among GPS coordinates and the genetic distance matrix as a matrix of pairwise allelic differences among genotypes.

2.6 | Environmental suitability, genetic rarity and spatiotemporal variation in phenotypic traits

We estimated an environmental suitability index for the *A. thaliana* population with an intensity function of a point process determined by 1093 *A. thaliana* individuals. The intensity function of a point process represents mathematically a collection of points in a given space. To this end, we used the 'density.ppp' function of the `R` package `spatstat` v.2.2-0 (Baddeley et al., 2015) to calculate a Gaussian kernel with a smoothing sigma of 10 m and without edge correction. We compared outputs with larger sigma values, but all the analyses were consistent (results not shown). Areas with several individuals sampled over the sampling years (2012–2021) had high environmental suitability indexes, whereas the opposite yielded to lower environmental suitability values. Environmental suitability results were scaled between 0 and 1.

We estimated genetic rarity for each *A. thaliana* individual using the genetic rarity index (Browne & Karubian, 2016) given by:

$$\text{Genetic rarity index} = \sum_{l=1}^{L} \left(p_a + p_b\right)_l / 2L, \tag{1}$$

where L is the total number of loci, and p_a and p_b are the frequencies of alleles a and b at locus l in the population. Individuals with lower values of the genetic rarity index contain rare, low-frequency alleles compared to individuals harbouring relatively common, higher frequency alleles (Browne & Karubian, 2016). Based on the spatial location of genotyped individuals, we used ordinary kriging interpolation at 1 m resolution to calculate a surface of the spatial distribution of genetic rarity across the *A. thaliana* population with the `R` package `gstat` v.2.0-7 (Gräler et al., 2016) and an automatically calculated variogram with the `R` package `automap` v.1.0-14 (Hiemstra et al., 2009). The final interpolated surface was scaled between 0 and 1.

The relationship between environmental suitability and life-history traits and fitness was explored using simultaneous autoregressive (SAR) models with `SAM` to incorporate the spatial autocorrelation of data. We also used SAR to evaluate the relationship between genetic rarity and life-history traits and fitness.

3 | RESULTS

3.1 | Weather and sensor data

Weather records from the closest automatic meteorological stations to the *A. thaliana* population indicated slight differences in mean temperatures among years (Figure S1). The largest difference in minimum and maximum mean temperatures were of 1.1°C and 2.4°C, respectively. The warmest year was 2017. Precipitations were more variable among years, with several years between 250 and 300 mm of total rain. Two years exhibited peaks of rain above 400 (2018) and 550 mm (2013). Data loggers deployed at different vegetation types did not show pronounced differences in temperature among vegetation types over 2 years (Figure S2). The minimum and maximum daily temperatures (−10.4°C and 58.1°C, respectively) were recorded at locations dominated by scrubland. The largest difference in minimum and maximum mean temperatures were of 3.2°C and 6.2°C, respectively, and were also recorded from data loggers deployed in scrubland.

3.2 | Spatiotemporal variation in life-history traits and fitness

We estimated the effects of vegetation type, year and maternal line on variation in life-history traits and fitness of 279 *A. thaliana* maternal lines in a common garden experiment. Overall, the results showed that vegetation type did not significantly affect any trait (Table 1), whereas temporal variation was significant in all traits except flowering time (Table 1). Among-maternal line variation was significant for seed weight, recruitment and flowering time, but not for survival, fecundity and fitness (Table 1). We found quite consistent trait–trait correlation patterns for maternal lines collected at different years. For example, maternal lines with earlier flowering showed a trend for higher recruitment, survival and fecundity, and therefore higher fitness (Figure S5).

For each year, we estimated broad-sense heritability values (h^2) to assess the degree of genetic determination of each life-history trait and fitness. Consistently among years, those traits with significant among-maternal line variation also exhibited h^2 values different from zero (Table 1; Table S2). Seed weight was the trait with the highest h^2 values in all years, followed by flowering time and recruitment (Table 1; Table S2).

To examine how natural selection acted upon *A. thaliana* phenotypes from different years of collection grown in the common garden experiment, we estimated linear and quadratic selection gradients (β and γ) and selection differentials (s and C) for recruitment and flowering time using a GAM-based approach. The linear selection gradients and differentials were significant for flowering time in each set of maternal lines from each year of collection (Table S3). All linear selection gradients for flowering time were negative, suggesting that selection favoured *A. thaliana* individuals with earlier flowering in the environmental conditions of the common garden experiment. In contrast, only a few linear positive selection differentials were significant for recruitment. Finally, quadratic selection exhibited a non-significant pattern for recruitment and flowering time, indicating a minimal role of stabilizing or disruptive selection on these *A. thaliana* traits in the conditions during the common garden experiment.

To determine the distribution of genetic variation across space, we estimated Moran's I autocorrelation coefficients for the life-history traits and fitness in each year. In 2012 and 2015, none of Moran's I were significant for any trait (Table S4). In 2013, we detected the greatest spatial structure as recruitment, flowering time and survival exhibited significant Moran's I values (Table S4). The maximum distance intervals at which there was significant spatial relationships among individuals in 2013 ranged between 17 m for survival and 38 m for recruitment. In 2015, seed weight was the only trait with significant Moran's I values with the first distance interval of 4 m showing a significant spatial relationship (Table S4).

TABLE 1 Linear Mixed Models (LMMS) and Generalized LMMS (GLMMs) testing the effect of vegetation type, year and maternal line nested within year on life-history traits and fitness for 279 *Arabidopsis thaliana* maternal lines from the study population. Factors were tested through likelihood-ratio tests. Conditional or marginal R^2 for each factor and variable are given. The range of broad-sense heritability (h^2) among years are given. Descriptive statistics using individual means (±SE) and ranges for each variable are shown. Asterisks indicate significance: $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$, ns; non-significant

Variable	Vegetation type		Year		Maternal line		Range h^2	Mean (±SE)	Range var.	Units
	χ^2	R^2	χ^2	R^2	χ^2	R^2				
Seed weight	0.22 ns	0.001	9.90**	0.059	977.30***	0.851	0.87–0.93	1.07 ± 0.01	0.70–1.52	$\times 10^{-5}$ g
Recruitment	1.62 ns	0.005	29.27***	0.122	2307.70***	0.652	0.22–0.45	0.37 ± 0.01	0.04–0.69	Proportion
Flowering time	3.38 ns	0.008	0.85 ns	0.007	289.46***	0.387	0.26–0.44	103.8 ± 0.2	94.2–110.7	No. days
Survival	1.63 ns	0.002	19.29***	0.026	1.37 ns	0.026	0.00–0.11	0.68 ± 0.01	0.28–0.92	Proportion
Fecundity	2.27 ns	0.001	34.39***	0.023	0.01 ns	0.133	0.00–0.05	56.2 ± 1.4	16.4–137.8	Seeds/ind.
Fitness	1.60 ns	0.002	18.68***	0.024	0.46 ns	0.014	0.00–0.06	42.5 ± 1.2	6.8–115.5	Seeds/ind.

3.3 | Spatiotemporal variation at the molecular level

We genotyped 884 *A. thaliana* individuals sampled over 9 years (2012–2020) with 12 nuclear microsatellites to estimate spatiotemporal patterns of variation in genetic diversity, the fine-scale genetic structure and the influence of ecological- and genetic-related parameters on phenotypic variation. Up to 863 individuals grouped into 411 different MLGs. Based on the criterion to discriminate among different MLG, the 21 remaining individuals could not be assigned to any MLG. We found 280 of 411 MLG (68.1%) including one individual only (i.e. unique MLG). The remaining 131 MLG had multiple individuals per MLG.

Neither year ($\chi^2 = 5.71$, $p = 0.68$) nor vegetation type ($\chi^2 = 0.15$, $p = 0.93$) differed in the number of unique MLG (Figure 2a). The rest of MLG included between 2 and up to 67 individuals (mean ± SD = 4.5 ± 6.4 individuals per MLG) and occurred indistinctly in all years and vegetation types (Figure 2a). Most of non-unique MLG had either two (58 MLG) or three (28 MLG) individuals per MLG. In contrast, MLG with more than three individuals were rare. For instance, only two MLG included up to 21 and 67 identical individuals. The *A. thaliana* population exhibited slight oscillations in the Shannon–Wiener diversity index among years (Figure 2b) and among the three major vegetation types with *A. thaliana* (Figure 2c).

Expected heterozygosity (H_S) was similar among years (mean ± SD = 0.65 ± 0.02; range = 0.62–0.68) and vegetation types (mean ± SD = 0.70 ± 0.04; range = 0.62–0.71). The same occurred with observed heterozygosity (H_O) among years (mean ± SD = 0.04 ± 0.02; range = 0.01–0.07) and vegetation types (mean ± SD = 0.03 ± 0.01; range = 0.02–0.05). In fact, we did not detect significant differences in H_S and H_O among years and vegetation types ($p > 0.17$ in all cases). The molecular variance explained by years and vegetation types was of 1%, whereas individuals accounted for up to 94% of the variance in both analyses.

We detected significant positive fine-scale spatial genetic structure (FSGS) in all years (Figure 3; Figure S6), particularly within the first distance class (25 m) where mean kinship ranged between 0.042 and 0.105 (Figure 3; Figure S6). We also detected positive FSGS at different distance classes always below 100 m with different combinations in different years (Figure 3; Figure S6). The strength of the FSGS was also variable among years, with *Sp* values ranging from a low of 0.02 in 2020 to a high of 0.06 in 2015 (Figure 3; Figure S6). As far as the spatial distribution of heterozygous individuals across the *A. thaliana* population is concerned, we did not detect any spatial pattern significantly different from the null model (Figure S7).

3.4 | Ecological and genetic drivers of phenotypic variation

We conducted Mantel tests to determine whether variation in life-history traits and fitness of *A. thaliana* was related to the spatial location of individuals and/or to the genetic relatedness among

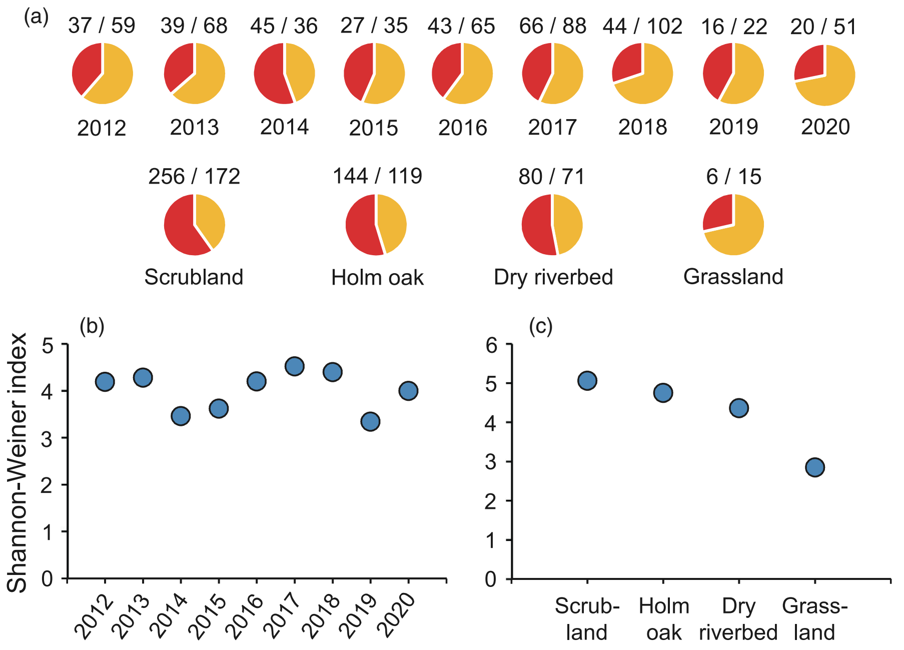

FIGURE 2 Genetic diversity parameters in *Arabidopsis thaliana* individuals from the study population across years and vegetation types. (a) Distribution of multilocus genotypes (MLGs) with one individual (orange) and MLGs with more than one individual (red) across years and vegetation types. (b) Shannon–Wiener diversity index for each year. (c) Shannon–Wiener diversity index for each vegetation type. The lowest value of the diversity index for grassland was due to the low sample size in this vegetation type.

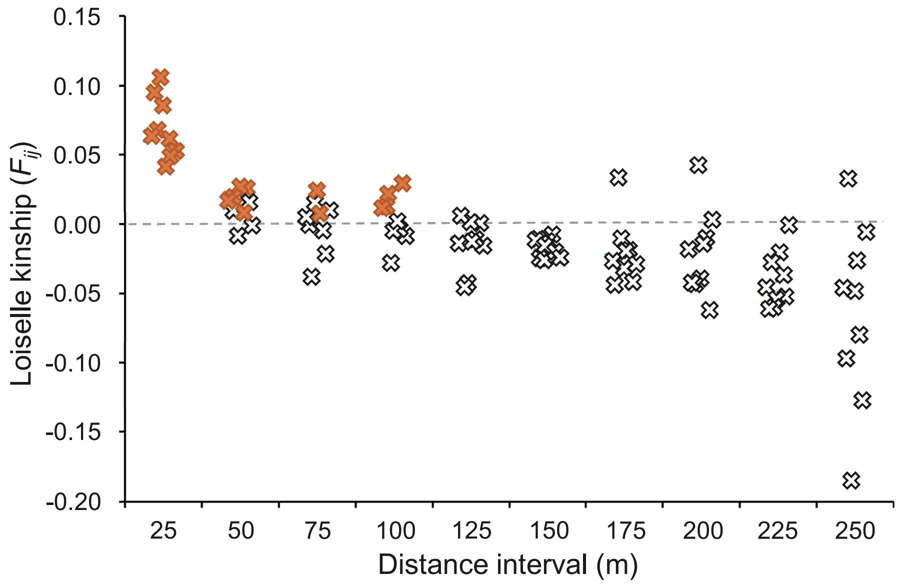

FIGURE 3 Jitter plot with the summary of the fine-scale spatial genetic structure at the *Arabidopsis thaliana* population. Spatial autocorrelation analyses used the Loiselle kinship coefficient (F_{ij}) and 10 distance classes for each year. Orange and white crosses indicate significant and non-significant Loiselle kinship coefficients, respectively (see text and Figure S6 for further details).

individuals. Genetic similarity among individuals, rather than their geographical distance, mostly accounted for differentiation in life-history traits and fitness among *A. thaliana* individuals (Table 2). However, in 2013, we found stronger significant relationships between phenotypic, geographical and genetic distances (Table 2). Except in 2013, we found significant Mantel tests in traits with higher broad-sense heritability values, such as seed weight, recruitment and flowering time (Table 2). Finally, geographical and genetic distances were not independent from each other ($r = 0.12$–0.17; $p < 0.009$ in all cases; Mantel tests) except in 2015 ($r = -0.02$; $p = 0.79$; Mantel test).

Based on the entire dataset of *A. thaliana* individuals sampled ($N = 1093$) and genotyped ($N = 884$), we estimated the spatial distribution of environmental suitability and genetic rarity across the *A. thaliana* population (Figure 4), which were not significantly correlated ($r = 0.11$, $p = 0.15$; Dutilleul's *t*-test). We conducted SAR models to test the effects of environmental suitability and genetic rarity heterogeneity on variation in life-history traits and fitness. We found that heterogeneity in environmental suitability did not account for variation in any life-history trait and fitness (Table 3). In contrast, genetic rarity significantly correlated with several life-history traits and fitness, particularly in 2013 and 2014 (Table 3). When significant, individuals harbouring rare, low-frequency alleles produced lighter seeds, flowered earlier and exhibited increased recruitment, survival and fitness (Table 3).

4 | DISCUSSION

Understanding the ecological and evolutionary consequences of spatiotemporal environmental heterogeneity in population

TABLE 2 Mantel tests comparing phenotypic distance with geographic and genetic distances for life-history traits and fitness of 274 *Arabidopsis thaliana* maternal lines from the study population. Correlation coefficients for each sampling year are given. Significant coefficients in boldface. Asterisks indicate significance: ***$p < 0.001$, **$p < 0.01$, ns; non-significant

	2012		2013		2014		2015	
Variable	Geographical	Genetic	Geographical	Genetic	Geographical	Genetic	Geographical	Genetic
Seed weight	0.01 ns	**0.28*** **	0.04 ns	0.02 ns	0.01 ns	**0.20*** **	**0.17** **	**0.28*** **
Recruitment	−0.02 ns	0.03 ns	**0.13** **	**0.29*** **	0.01 ns	0.06 ns	−0.03 ns	**0.17** **
Flowering time	−0.01 ns	0.04 ns	**0.14** **	**0.20** **	−0.06 ns	**0.31** **	−0.02 ns	0.12 ns
Survival	−0.03 ns	0.01 ns	**0.13** **	**0.12** **	0.01 ns	0.01 ns	−0.03 ns	0.09 ns
Fecundity	−0.04 ns	−0.02 ns	0.04 ns	−0.06 ns	−0.05 ns	0.03 ns	−0.03 ns	0.09 ns
Fitness	−0.10 ns	−0.01 ns	**0.11** **	−0.05 ns	−0.03 ns	0.05 ns	−0.04 ns	0.12 ns

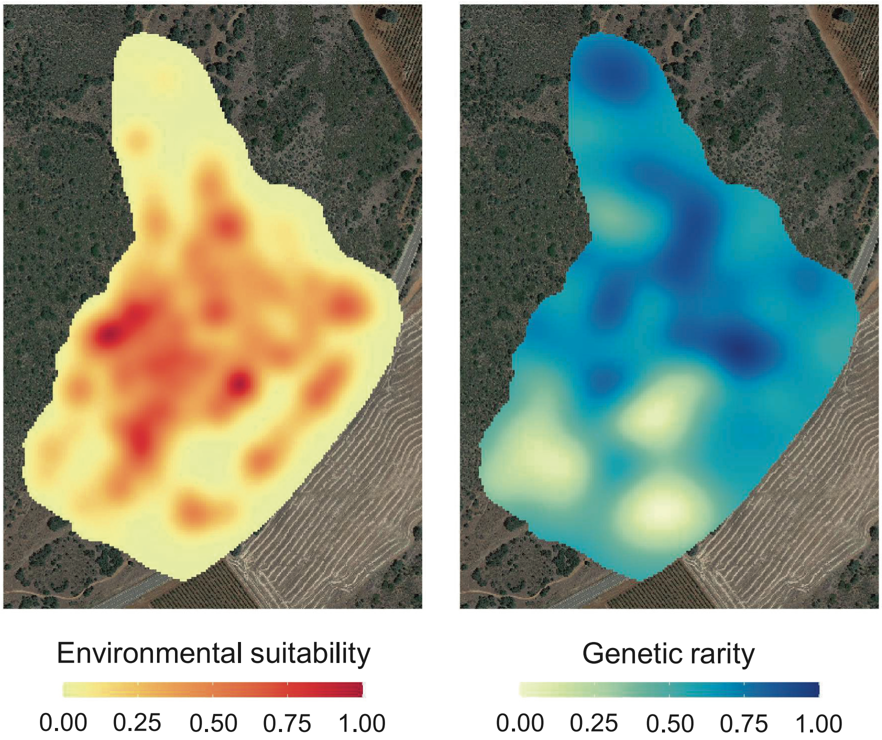

FIGURE 4 Spatial distribution of environmental suitability (left) and genetic rarity (right) for *Arabidopsis thaliana* across the study population (see text for details).

performance requires the comprehension of the forces that shape and maintain genetic variation within populations. Given the increasing role of *Arabidopsis thaliana* as a model system for ecology and evolution (Takou et al., 2019), within-population studies in *A. thaliana* increase our knowledge on its local-scale phenotypic variation (Brachi et al., 2013; Frachon et al., 2017; Gómez et al., 2018; Handley et al., 2005; Kuittinen et al., 1997; Méndez-Vigo et al., 2013) and spatiotemporal heterogeneity effects on fitness-related traits (Bell & Lechowicz, 1991; Stratton & Bennington, 1996; Frachon et al., 2017; Gómez et al., 2018). Here, we combined long-term field sampling, experimental and molecular approaches at a fine spatial scale to disentangle the effects of environmental heterogeneity on quantitative and molecular variation in a natural *A. thaliana* population.

The results showed non-significant spatial heterogeneity in *A. thaliana* quantitative traits (Table 1). Previous studies on other *A. thaliana* populations, however, detected local-scale environmental heterogeneity accounting for within-population quantitative genetic variation (Bell & Lechowicz, 1991; Stratton & Bennington, 1996; Frachon et al., 2017). We believe that *A. thaliana* at the Moral de Calatrava population also responds to spatial heterogeneity, but vegetation type was probably too coarse to detect a significant effect. The similar temperature records from data loggers deployed at different vegetation types suggest that vegetation type per se is not a good indicator of the ecological factors accounting for spatial heterogeneity in *A. thaliana*. Thus, further research on individual-scale attributes, such as soil properties and plant neighbourhood (Frachon et al., 2017, 2019), may help detect the effects of finer-scale heterogeneity on variation in life-history traits.

In contrast, we found that, except for flowering time, all traits varied across years (Table 1). Although weather records did not

TABLE 3 Simultaneous autoregressive (SAR) models testing the effects of environmental suitability and genetic rarity on life-history traits and fitness of 274 *Arabidopsis thaliana* maternal lines from the study population. Correlation coefficients for each sampling year are given. Significant coefficients in boldface. Asterisks indicate significance: ***$p < 0.001$, **$p < 0.01$, ns; non-significant

	2012		2013		2014		2015	
Variable	Suitability	Rarity	Suitability	Rarity	Suitability	Rarity	Suitability	Rarity
Seed weight	−0.01 ns	0.12 ns	0.09 ns	0.07 ns	−0.12 ns	**0.60*** **	−0.05 ns	**0.59*** **
Recruitment	0.04 ns	−0.01 ns	−0.08 ns	**−0.39*** **	−0.08 ns	**−0.31** **	0.02 ns	0.21 ns
Flowering time	0.05 ns	0.21 ns	0.11 ns	**0.44*** **	0.04 ns	**0.66*** **	−0.02 ns	0.10 ns
Survival	0.01 ns	−0.23 ns	−0.06 ns	**−0.42*** **	−0.03 ns	−0.28 ns	0.01 ns	0.09 ns
Fecundity	0.06 ns	−0.14 ns	−0.06 ns	−0.02 ns	−0.03 ns	**−0.32** **	0.05 ns	0.04 ns
Fitness	0.04 ns	−0.20 ns	−0.07 ns	**−0.31** **	−0.03 ns	**−0.38*** **	0.02 ns	0.09 ns

suffice to clearly identify the source of year-to-year variation, every sampling year was different, determining the number and location of *A. thaliana* individuals (Figure 1b). Based on our experience, the critical events for *A. thaliana* are a dry autumn, when the peak of germination occurs in Iberian *A. thaliana* (Montesinos et al., 2009; Picó, 2012), and a dry late winter, when floral induction and first flowering take place. Both scenarios may reduce survival and fecundity to a large extent. Nonetheless, natural *A. thaliana* populations in several Iberian environments are made of cohorts of autumn- and spring-germinated individuals (Montesinos et al., 2009; Picó, 2012) emerging from the transient seed bank (Falahati-Anbaran et al., 2014; Lundemo et al., 2009; Montesinos et al., 2009; Postma et al., 2016), meaning that the species has the potential to buffer environmental stochasticity by recruiting new individuals at different times of the year. We also detected different cohorts of individuals at the Moral de Calatrava population (F.X. Picó and C. Alonso-Blanco, pers. obs.), which will be considered in forthcoming studies to better understand the patterns and drivers of temporal fluctuation in *A. thaliana* abundance.

Our variance decomposition analysis also revealed those traits with a higher degree of genetic determination: seed weight, recruitment and flowering time (Table 1 and Table S2). However, in the environmental conditions in which the common garden experiment was conducted, selection acted upon flowering time (i.e. earlier flowering maternal lines significantly showed higher fitness) in each set of maternal lines collected over 4 years (Table S3). In addition, flowering time was the only trait showing no variation among maternal lines from each sampling year (Table 1), suggesting that the genetic component of flowering time variation was higher than that of the rest of traits analysed. The detection of selection favouring early flowering in *A. thaliana* is not rare in the literature, as it has also been found at regional scales using accessions (Exposito-Alonso et al., 2018; Kenney et al., 2014). Although the intensity and direction of selection can vary over time, accounting for the maintenance of genetic variation in flowering time within *A. thaliana* populations (Ågren et al., 2017; Exposito-Alonso et al., 2018; Fournier-Level et al., 2013; Manzano-Piedras et al., 2014; Taylor et al., 2017), the concordance between large- and local-scale results (i.e. accession- vs. population-based approaches) provides additional evidence that *A. thaliana* becomes locally adapted to varying environments by adjusting the seasonal timing of key developmental events of life-cycle phenology (Burghardt et al., 2016; Chiang et al., 2013; Debieu et al., 2013; Donohue, 2005; Huang et al., 2010; Marcer et al., 2018; Vidigal et al., 2016; Wilczek et al., 2009).

From a molecular viewpoint, *A. thaliana* has a large genetic pool at the study population, as shown in other *A. thaliana* populations (Bakker et al., 2006; Bomblies et al., 2010; Castilla et al., 2020; Falahati-Anbaran et al., 2014; Frachon et al., 2017; Gomaa et al., 2011; Gómez et al., 2018; Méndez-Vigo et al., 2013). In particular, almost half of the genotyped individuals were different MLGs and nearly 70% of them were represented by one individual. This result suggests two properties of *A. thaliana* at the study population. On the one hand, the high genetic diversity detected is probably the result of the old history of the population. The lack of vegetation type and year effects on genetic diversity and the low amount of genetic differentiation accounted for by vegetation type and year also supported the notion of population maturity. On top of that, the *A. thaliana* population is classified as an Iberian relict population (Castilla et al., 2020). This means that the study population, along with other nearby *A. thaliana* populations (Castilla et al., 2020; Tabas-Madrid et al., 2018), belongs to a genetic lineage with a long evolutionary history (Durvasula et al., 2017). Furthermore, it has be shown that Iberian and Moroccan *A. thaliana* relicts tend to occur in areas with more stable vegetation dynamics since the Last Glacial Maximum and during the Holocene (Toledo et al., 2020). Overall, we believe that *A. thaliana* has had the time to generate and distribute genetic diversity across the whole area including the *A. thaliana* population of study over many generations.

On the other hand, the high proportion of MLGs represented by one individual at the *A. thaliana* population also stresses the potential of *A. thaliana* to generate new genetic variants, probably accounted for by low but steady outcrossing rates. In fact, *A. thaliana* populations may exhibit local outcrossing hot spots (Bomblies et al., 2010), which have the potential to reshuffle genetic variants and therefore boost genetic diversity. In the *A. thaliana* population of study, we did not detect such outcrossing hot spots, based on the non-significant effect of vegetation type and year on heterozygosity and the lack of spatial structure for heterozygous individuals

(Figure S7). Nevertheless, we observed small bees and hoverflies visiting *A. thaliana* and congeners in dense flowering patches or in particular years when *A. thaliana* becomes ubiquitous at the study population (F.X. Picó, pers. obs.).

Despite the limited effect of vegetation type and year on variation in quantitative traits and genetic diversity, *A. thaliana* is genetically structured at the study population. We found a consistent significant fine-scale genetic structure at short distances (Figure 3; Figure S6), which varied in distance and intensity across years. Furthermore, such fine-scale genetic structure was a putatively good predictor of phenotypic differentiation in *A. thaliana*, as phenotypic distance exhibited stronger significant relationships with genetic relatedness than with geographical distance (Table 2), and also supported by the weak spatial autocorrelation of traits (Table S4). The exception was 2013, which was also the wettest year of the series (particularly in autumn and spring; Figure S1). In 2013, we captured significant spatial autocorrelations and relationships between phenotypic distance and geographical and genetic distances for several traits (Table 2; Table S4). This result stresses the need of repeated sampling over time to extract realistic population attributes depicting the complexity of natural *A. thaliana* populations (Frachon et al., 2017; Gomaa et al., 2011; Gómez et al., 2018). We conclude that the role of biotic vectors promoting dispersal (e.g. wild boars removing the soil) may be unimportant at the study population and that phenotypic variation seems to be chiefly driven by the known limited *A. thaliana*'s dispersal ability and its self-fertilizing habit (Bomblies et al., 2010; Castilla et al., 2020; Falahati-Anbaran et al., 2014; Lundemo et al., 2009).

As a result of our long-term field survey over a decade, we were able to estimate the spatial location of hot spots and cold spots of environmental suitability and genetic rarity for *A. thaliana* across the study population (Figure 4). Genetic rarity emerged as a good predictor of phenotypic variation, whereas environmental suitability was not significantly related to any phenotypic trait (Table 3). When significant, individuals with lower genetic rarity (harbouring rarer alleles) exhibited early flowering. Based on our results, we hypothesise that the appearance of novel earlier flowering phenotypes would imply a lower allelic frequency in the population. This is plausible because low-frequency nucleotide polymorphisms in flowering genes normally imply loss- or change-of-function alleles, which are related to early-flowering phenotypes (Alonso-Blanco et al., 2009; Fulgione et al., 2022; Johanson et al., 2000; Méndez-Vigo et al., 2011). Further research is needed to better understand the effect of selection on rare genotypes and the relationship between low-frequency neutral and functional alleles. In any case, our results stress the preponderance of genetic attributes over ecological factors driving phenotypic variation in *A. thaliana* at the Moral de Calatrava population.

This study also lays the foundations for detecting within-population variation in the genetic basis of evolutionarily important traits in *A. thaliana*. For example, it will be important in forthcoming population-based studies to quantify the frequency of functional variants for key flowering genes, which most of them are rare at a worldwide scale but can be common at the local scale (Alonso-Blanco et al., 2009; Brachi et al., 2013; Gould & Stinchcombe, 2017; Le Corre, 2005; Méndez-Vigo et al., 2011; Sánchez-Bermejo et al., 2012). Furthermore, pleiotropic effects of developmental genes on other plant traits seem to be the rule (Auge et al., 2019; Blair et al., 2017; Chiang et al., 2009, 2013; Donohue, 2014; Hanemian et al., 2020; Lovell et al., 2013; McKay et al., 2003; Scarcelli et al., 2007), providing the mechanism to understand the tight association found among life-history traits in *A. thaliana*. Overall, we believe that the increase in the number of extensive within-population studies, across space and over time, from contrasting environments will provide a missing piece to build realistic mechanistic models of evolutionary change in *A. thaliana*.

AUTHOR CONTRIBUTIONS

F. Xavier Picó and Carlos Alonso-Blanco conceived the idea. Belén Méndez-Vigo, Carlos Alonso-Blanco, Rocío Gómez, Arnald Marcer and F. Xavier Picó generated materials and data. Antonio R. Castilla and F. Xavier Picó analysed the data. F. Xavier Picó wrote the first version of the manuscript and all authors contributed to it.

ACKNOWLEDGEMENTS

We thank David Ragel, Mercedes Ramiro, María Amores, Moises Exposito-Alonso, Marisol Muñoz and the staff of the El Castillejo Botanical Garden for field, laboratory and/or greenhouse assistance. The Laboratorio de Ecología Molecular from Estación Biológica de Doñana and the Doñana's Singular Scientific and Technical Infrastructure (ICTS-RBD) provided logistical support. We thank the anonymous reviewers for their suggestions and comments on an earlier version of this manuscript. We are grateful to Agencia Estatal de Meteorología of Spain (AEMET) for providing meteorological data. We also thank the administration of the Sierra de Grazalema for permission to work at the El Castillejo Botanical Garden. This research was funded by grants CGL2016-77720-P and PID2019-104135GB-I00 (AEI/10.13039/501100011033) from the Agencia Estatal de Investigación (AEI) of Spain and the Fondo Europeo de Desarrollo Regional (FEDER, UE) to F.X.P.

CONFLICT OF INTEREST

The authors have no conflict of interest to declare.

PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1111/1365-2745.13981.

DATA AVAILABILITY STATEMENT

Raw data are available from Dryad Digital Repository https://doi.org/10.5061/dryad.7sqv9s4vt (Picó, 2022).

ORCID

Belén Méndez-Vigo https://orcid.org/0000-0002-9850-536X
Antonio R. Castilla https://orcid.org/0000-0002-4124-738X

Rocío Gómez https://orcid.org/0000-0002-8218-7838
Arnald Marcer https://orcid.org/0000-0002-6532-7712
Carlos Alonso-Blanco https://orcid.org/0000-0002-4738-5556
F. Xavier Picó https://orcid.org/0000-0003-2849-4922

REFERENCES

Ågren, J., Oakley, C. G., Lundemo, S., & Schemske, D. W. (2017). Adaptive divergence in flowering time among natural populations of *Arabidopsis thaliana*: Estimates of selection and QTL mapping. *Evolution*, *71*, 550–564.

Ågren, J., Oakley, C. G., McKay, J. K., Lovell, J. T., & Schemske, D. W. (2013). Genetic mapping of adaptation reveals fitness tradeoffs in *Arabidopsis thaliana*. *Proceedings of the National Academy of Sciences of the United States of America*, *110*, 21077–21082.

Alonso-Blanco, C., Aarts, M. G. M., Bentsink, L., Keurentjes, J. J. B., Reymond, M., Vreugdenhil, D., & Koornneef, M. (2009). What has natural variation taught us about plant development, physiology, and adaptation? *The Plant Cell*, *21*, 1877–1896.

Argyres, A. Z., & Schmitt, J. (1991). Microgeographic genetic structure of morphological and life history traits in a natural population of *Impatiens capensis*. *Evolution*, *45*, 175–189.

Atwell, S., Huang, Y. S., Vilhjálmsson, B. J., Willems, G., Horton, M., Li, Y., Meng, D., Platt, A., Tarone, A. M., Hu, T. T., Jiang, R., Muliyati, N. W., Zhang, X., Amer, M. A., Baxter, I., Brachi, B., Chory, J., Dean, C., Debieu, M., … Nordborg, M. (2010). Genome-wide association study of 107 phenotypes in *Arabidopsis thaliana* inbred lines. *Nature*, *465*, 627–631.

Auge, G. A., Penfield, S., & Donohue, K. (2019). Pleiotropy in developmental regulation by flowering-pathway genes: Is it an evolutionary constraint? *New Phytologist*, *224*, 55–70.

Baddeley, A., Rubak, E., & Turner, R. (2015). *Spatial point patterns: Methodology and applications with R*. Chapman and Hall/CRC Press.

Bakker, E. G., Stahl, E. A., Toomajan, C., Nordborg, M., Kreitman, M., & Bergelson, J. (2006). Distribution of genetic variation within and among local populations of *Arabidopsis thaliana* over its species range. *Molecular Ecology*, *15*, 1405–1418.

Barrett, R. D. H., & Schluter, D. (2008). Adaptation from standing genetic variation. *Trends in Ecology and Evolution*, *23*, 38–44.

Barton, N., & Keightley, P. (2002). Understanding quantitative genetic variation. *Nature Reviews Genetics*, *3*, 11–21.

Barton, N. H., & Turelli, M. (1989). Evolutionary quantitative genetics: How little do we know? *Annual Review of Genetics*, *23*, 337–370.

Bates, D., Mächler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statistical Software*, *67*, 1–48.

Bell, G., & Lechowicz, M. J. (1991). The ecology and genetics of fitness in forest plants. I. Environmental heterogeneity measured by explant trials. *Journal of Ecology*, *79*, 663–685.

Bentsink, L., Hanson, J., Hanhart, C. J., Blankestijn-de Vries, H., Coltrane, C., Keizer, P., El-Lithy, M., Alonso-Blanco, C., de Andrés, M. T., Reymond, M., van Eeuwijk, F., Smeekens, S., & Koornneef, M. (2010). Natural variation for seed dormancy in Arabidopsis is regulated by additive genetic and molecular pathways. *Proceedings of the National Academy of Sciences of the United States of America*, *107*, 4264–4289.

Blair, L., Auge, G., & Donohue, K. (2017). Effect of *FLOWERING LOCUS C* on seed germination depends on dormancy. *Functional Plant Biology*, *44*, 493–506.

Bomblies, K., Yant, L., Laitinen, R. A., Kim, S.-T., Hollister, J. D., Warthmann, N., Fitz, J., & Weigel, D. (2010). Local-scale patterns of genetic variability, outcrossing, and spatial structure in natural stands of *Arabidopsis thaliana*. *PLoS Genetics*, *6*, e1000890.

Brachi, B., Faure, N., Horton, M., Flahauw, E., Vazquez, A., Nordborg, M., Bergelson, J., Cuguen, J., & Roux, F. (2010). Linkage and association mapping of *Arabidopsis thaliana* flowering time in nature. *PLoS Genetics*, *6*, e1000940.

Brachi, B., Villoutreix, R., Faure, N., Hautekèete, N., Piquot, Y., Pauwels, M., Roby, D., Cuguen, J., Bergelson, J., & Roux, F. (2013). Investigation of the geographical scale of adaptive phenological variation and its underlying genetics in *Arabidopsis thaliana*. *Molecular Ecology*, *22*, 4222–4240.

Browne, L., & Karubian, J. (2016). Frequency-dependent selection for rare genotypes promotes genetic diversity of a tropical palm. *Ecology Letters*, *19*, 1439–1447.

Burghardt, L. T., Edwards, B. R., & Donohue, K. (2016). Multiple paths to similar germination behavior in *Arabidopsis thaliana*. *New Phytologist*, *209*, 1301–1312.

Castilla, A. R., Garrote, P. J., Żywiec, M., Calvo, G., Suárez-Esteban, A., Delibes, M., Godoy, J. A., Picó, F. X., & Fedriani, J. M. (2019). Genetic rescue by distant trees mitigates qualitative pollen limitation imposed by fine-scale spatial genetic structure. *Molecular Ecology*, *28*, 4363–4374.

Castilla, A. R., Méndez-Vigo, B., Marcer, A., Martínez-Minaya, J., Conesa, D., Picó, F. X., & Alonso-Blanco, C. (2020). Ecological, genetic and evolutionary drivers of regional genetic differentiation in *Arabidopsis thaliana*. *BMC Evolutionary Biology*, *20*, 71.

Chiang, G. C. K., Barua, D., Dittmar, E., Kramer, E. M., Amasino, R. M., & Donohue, K. (2009). Major flowering time gene, *FLOWERING LOCUS C*, regulates seed germination in *Arabidopsis thaliana*. *Proceedings of the National Academy of Sciences of the United States of America*, *106*, 11661–11666.

Chiang, G. C. K., Barua, D., Dittmar, E., Kramer, E. M., Rubio de Casas, R., & Donohue, K. (2013). Pleiotropy in the wild: The dormancy gene *DOG1* exerts cascading control on life cycles. *Evolution*, *67*, 883–893.

De Frenne, P., Rodríguez-Sánchez, F., Coomes, D. A., Baeten, L., Verstraeten, G., Vellend, M., Bernhardt-Römermann, M., Brown, C. D., Brunet, J., Cornelis, J., Decocq, G. M., Dierschke, H., Eriksson, O., Gilliam, F. S., Hédl, R., Heinken, T., Hermy, M., Hommel, P., Jenkins, M. A., … Verheyen, K. (2013). Microclimate moderates plant responses to macroclimate warming. *Proceedings of the National Academy of Sciences of the United States of America*, *110*, 18561–18565.

Debieu, M., Tang, C., Stich, B., Sikosek, T., Effgen, S., Josephs, E., Schmitt, J., Nordborg, M., Koornneef, M., & de Meaux, J. (2013). Co-variation between seed dormancy, growth rate and flowering time changes with latitude in *Arabidopsis thaliana*. *PLoS ONE*, *8*, e61075.

Denney, D. A., Jameel, M. I., Bemmels, J. B., Rochford, M. E., & Anderson, J. T. (2020). Small spaces, big impacts: Contributions of micro-environmental variation to population persistence under climate change. *AoB PLANTS*, *12*, plaa005.

Donohue, K. (2005). Niche construction through phenological plasticity: Life history dynamics and ecological consequences. *New Phytologist*, *166*, 83–92.

Donohue, K. (2014). Why ontogeny matters during adaptation: Developmental niche construction and pleiotropy across the life cycle in *Arabidopsis thaliana*. *Evolution*, *68*, 32–47.

Durvasula, A., Fulgione, A., Gutaker, R. M., Alacakaptan, S. I., Flood, P. J., Neto, C., Tsuchimatsu, T., Burbano, H. A., Picó, F. X., Alonso-Blanco, A., et al. (2017). African genomes illuminate the early history and transition to selfing in *Arabidopsis thaliana*. *Proceedings of the National Academy of Sciences of the United States of America*, *114*, 5213–5218.

Ehrlén, J., & Valdés, A. (2020). Climate drives among-year variation in natural selection on flowering time. *Ecology Letters*, *23*, 653–662.

El-Lithy, M. E., Clerkx, E. J., Ruys, G. J., Koornneef, M., & Vreugdenhil, D. (2004). Quantitative trait locus analysis of growth-related traits in a new Arabidopsis recombinant inbred population. *Plant Physiology*, *135*, 444–458.

Exposito-Alonso, M., 500 Genomes Field Experiment Team, Burbano, H. A., Bossdorf, O., Nielsen, R., & Weigel, D. (2019). Natural selection on the *Arabidopsis thaliana* genome in present and future climates. *Nature*, *573*, 126–129.

Exposito-Alonso, M., Brennan, A. C., Alonso-Blanco, C., & Picó, F. X. (2018). Spatio-temporal variation in fitness responses to contrasting environments in *Arabidopsis thaliana*. *Evolution*, *72*, 1570–1586.

Falahati-Anbaran, M., Lundemo, S., & Stenøien, H. K. (2014). Seed dispersal in time can counteract the effect of gene flow between natural populations of *Arabidopsis thaliana*. *New Phytologist*, *202*, 1043–1054.

Fournier-Level, A., Korte, A., Cooper, M. D., Nordborg, M., Schmitt, J., & Wilczek, A. M. (2011). A map of local adaptation in *Arabidopsis thaliana*. *Science*, *334*, 86–89.

Fournier-Level, A., Wilczek, A. M., Cooper, M. D., Roe, J. L., Anderson, J., Eaton, D., Moyers, B. T., Petipas, R. H., Schaeffer, R. N., Pieper, B., Reymond, M., Koornneef, M., Welch, S. M., Remington, D. L., & Schmitt, J. (2013). Paths to selection on life history loci in different natural environments across the native range of *Arabidopsis thaliana*. *Molecular Ecology*, *22*, 3552–3566.

Frachon, L., Libourel, C., Villoutreix, R., Carrère, S., Glorieux, C., Huard-Chauveau, C., Navascués, M., Gay, L., Vitalis, R., Baron, E., Amsellem, L., Bouchez, O., Vidal, M., le Corre, V., Roby, D., Bergelson, J., & Roux, F. (2017). Intermediate degrees of synergistic pleiotropy drive adaptive evolution in ecological time. *Nature Ecology and Evolution*, *1*, 1551–1561.

Frachon, L., Mayjonade, B., Bartoli, C., Hautekèete, N. C., & Roux, F. (2019). Adaptation to plant communities across the genome of *Arabidopsis thaliana*. *Molecular Biology and Evolution*, *36*, 1442–1456.

Franks, S. J., Sim, S., & Weis, A. E. (2007). Rapid evolution of flowering time by an annual plant in response to a climate fluctuation. *Proceedings of the National Academy of Sciences of the United States of America*, *104*, 1278–1282.

Friedman, J., Middleton, T. E., & Rubin, M. J. (2019). Environmental heterogeneity generates intrapopulation variation in life-history traits in an annual plant. *New Phytologist*, *224*, 1171–1183.

Fulgione, A., Neto, C., Elfarargi, A. F., Tergemina, E., Ansari, S., Göktay, M., Dinis, H., Döring, N., Flood, P. J., Rodriguez-Pacheco, S., Walden, M., Koch, M. A., Roux, F., Hermisson, J., & Hancock, A. M. (2022). Parallel reduction in flowering time from de novo mutations enable evolutionary rescue in colonizing lineages. *Nature Communications*, *13*, 1461.

Galloway, L. F. (1995). Response to natural environmental heterogeneity: Maternal effects and selection on life-history characters and plasticities in *Mimulus guttatus*. *Evolution*, *49*, 1095–1107.

Galpern, P., Manseau, M., Hettinga, P., Smith, K., & Wilson, P. (2012). Allelematch: An R package for identifying unique multilocus genotypes where genotyping error and missing data may be present. *Molecular Ecology Resources*, *12*, 771–778.

Gillespie, J. H., & Turelli, M. (1989). Genotype-environment interactions and the maintenance of polygenic variation. *Genetics*, *137*, 129–138.

Gomaa, N. H., Montesinos-Navarro, A., Alonso-Blanco, C., & Picó, F. X. (2011). Temporal variation in genetic diversity and effective population size of Mediterranean and subalpine *Arabidopsis thaliana* populations. *Molecular Ecology*, *20*, 3540–3554.

Gómez, R., Méndez-Vigo, B., Marcer, A., Alonso-Blanco, C., & Picó, F. X. (2018). Quantifying temporal change in plant population attributes: Insights from a resurrection approach. *AoB PLANTS*, *10*, ply063.

Gould, B. A., & Stinchcombe, J. R. (2017). Population genomic scans suggest novel genes underlie convergent flowering time evolution in the introduced range of *Arabidopsis thaliana*. *Molecular Ecology*, *26*, 92–106.

Gräler, B., Pebesma, E., & Heuvelink, G. (2016). Spatio-Temporal Interpolation using gstat. *The R Journal*, *8*, 204–218.

Hancock, A. M., Brachi, B., Faure, N., Horton, M. W., Jarymowycz, L. B., Sperone, F. G., Toomajian, C., Roux, F., & Bergelson, J. (2011). Adaptation to climate across the *Arabidopsis thaliana* genome. *Science*, *334*, 83–86.

Handley, R., Ekbom, B., & Ågren, J. (2005). Variation in trichome density and resistance against a specialist insect herbivore in natural populations of *Arabidopsis thaliana*. *Ecological Entomology*, *30*, 284–292.

Hanemian, M., Vasseur, F., Marchadier, E., Gilbault, E., Bresson, J., Gy, I., Violle, C., & Loudet, O. (2020). Natural variation at *FLM* splicing has pleiotropic effects modulating ecological strategies in *Arabidopsis thaliana*. *Nature Communications*, *11*, 4140.

Hardy, O. J., & Vekemans, X. (2002). SPAGeDi: A versatile computer program to analyse spatial genetic structure at the individual or population levels. *Molecular Ecology Notes*, *2*, 618–620.

Hiemstra, P. H., Pebesma, E. J., Twenhöfel, C. J. W., & Heuvelink, G. B. M. (2009). Real-time automatic interpolation of ambient gamma dose rates from the Dutch radioactivity monitoring network. *Computers and Geosciences*, *35*, 1711–1721.

Huang, X., Schmitt, J., Dorn, L., Griffith, C., Effgen, S., Takao, S., Koornneef, M., & Donohue, K. (2010). The earliest stages of adaptation in an experimental plant population: Strong selection on QTLS for seed dormancy. *Molecular Ecology*, *19*, 1335–1351.

Johanson, U., West, J., Lister, C., Michaels, S., Amasino, R., & Dean, C. (2000). Molecular analysis of *FRIGIDA*, a major determinant of natural variation in Arabidopsis flowering time. *Science*, *290*, 344–347.

Kalisz, S. (1986). Variable selection on the timing of germination in *Collinsia verna* (Scrophulariaceae). *Evolution*, *40*, 479–491.

Kenney, A. M., McKay, J. K., Richards, J. H., & Juenger, T. E. (2014). Direct and indirect selection on flowering time, water-use efficiency (WUE, δ^{13}C), and WUE plasticity to drought in *Arabidopsis thaliana*. *Ecology and Evolution*, *4*, 4505–4521.

Kuester, A., Wilson, A., Chang, S. M., & Baucom, R. S. (2016). A resurrection experiment finds evidence of both reduced genetic diversity and potential adaptive evolution in the agricultural weed *Ipomoea purpurea*. *Molecular Ecology*, *25*, 4508–4520.

Kuittinen, H., Mattila, A., & Savolainen, O. (1997). Genetic variation at marker loci and in quantitative traits in natural populations of *Arabidopsis thaliana*. *Heredity*, *79*, 144–152.

Le Corre, V. (2005). Variation at two flowering time genes within and among populations of *Arabidopsis thaliana*: Comparison with markers and traits. *Molecular Ecology*, *14*, 4181–4192.

Linhart, Y. B., & Grant, M. C. (1996). Evolutionary significance of local genetic differentiation in plants. *Annual Review of Ecology and Systematics*, *27*, 237–277.

Loiselle, B. A., Sork, V. L., Nason, J., & Graham, C. (1995). Spatial genetic structure of a tropical understory shrub, *Psychotria officinalis* (Rubiaceae). *American Journal of Botany*, *82*, 1420–1425.

Lovell, J. T., Juenger, T. E., Michaels, S. D., Lasky, J. R., Platt, A., Richards, J. H., Yu, X., Easlon, H. M., Sen, S., & McKay, J. K. (2013). Pleiotropy of *FRIGIDA* enhances the potential for multivariate adaptation. *Proceedings of the Royal Society B: Biological Sciences*, *280*, 20131043.

Lundemo, S., Falahati-Anbaran, M., & Stenøien, H. K. (2009). Seed banks cause elevated generation times and effective population sizes of *Arabidopsis thaliana* in northern Europe. *Molecular Ecology*, *18*, 2798–2811.

Manzano-Piedras, E., Marcer, A., Alonso-Blanco, C., & Picó, F. X. (2014). Deciphering the adjustment between environment and life history in annuals: Lessons from a geographically-explicit approach in *Arabidopsis thaliana*. *PLoS ONE*, *9*, e87836.

Marcer, A., Vidigal, D. S., James, P. M. A., Fortin, M.-J., Mendez-Vigo, B., Hilhorst, H. W. M., Bentsink, L., Alonso-Blanco, C., & Picó, F. X. (2018). Temperature fine-tunes Mediterranean *Arabidopsis thaliana* life-cycle phenology geographically. *Plant Biology*, *20*, 148–156.

Martínez-Berdeja, A., Stitzer, M. C., Taylor, M. A., Okada, M., Ezcurra, E., Runcie, D. E., & Schmitt, J. (2020). Functional variants of *DOG1* control seed chilling responses and variation in seasonal life-history strategies in *Arabidopsis thaliana*. *Proceedings of the*

National Academy of Sciences of the United States of America, *117*, 2526–2534.

McKay, J. K., Richards, J. H., & Mitchell-Olds, T. (2003). Genetics of drought adaptation in *Arabidopsis thaliana*: I. Pleiotropy contributes to genetic correlations among ecological traits. *Molecular Ecology*, *12*, 1137–1151.

McLaughlin, B. C., Ackerly, D. D., Klos, P. Z., Natali, J., Dawson, T. E., & Thompson, S. E. (2017). Hydrologic refugia, plants, and climate change. *Global Change Biology*, *23*, 2941–2961.

Méndez-Vigo, B., Gomaa, N. H., Alonso-Blanco, C., & Picó, F. X. (2013). Among- and within-population variation in flowering time of Iberian *Arabidopsis thaliana* estimated in field and glasshouse conditions. *New Phytologist*, *197*, 1332–1343.

Méndez-Vigo, B., Picó, F. X., Ramiro, M., Martínez-Zapater, J. M., & Alonso-Blanco, C. (2011). Altitudinal and climatic adaptation is mediated by flowering traits and *FRI*, *FLC*, and *PHYC* genes in Arabidopsis. *Plant Physiology*, *157*, 1942–1955.

Mitchell-Olds, T., & Bergelson, J. (1990). Statistical genetics of an annual plant, *Impatiens capensis*. I. Genetic basis of quantitative variation. *Genetics*, *124*, 407–415.

Mitchell-Olds, T., Willis, J. H., & Goldstein, D. B. (2007). Which evolutionary processes influence natural genetic variation for phenotypic traits? *Nature Reviews Genetics*, *8*, 845–856.

Mojica, J. P., Lee, Y. W., Willis, J. H., & Kelly, J. K. (2012). Spatially and temporally varying selection on intrapopulation quantitative trait loci for a life history trade-off in *Mimulus guttatus*. *Molecular Ecology*, *21*, 3718–3728.

Montesinos, A., Tonsor, S. J., Alonso-Blanco, C., & Picó, F. X. (2009). Demographic and genetic patterns of variation among populations of *Arabidopsis thaliana* from contrasting native environments. *PLoS ONE*, *4*, e7213.

Morrissey, M. B., & Sakrejda, K. (2013). Unification of regression-based methods for the analysis of natural selection. *Evolution*, *67*, 2094–2100.

Nakagawa, S., Johnson, P. C. D., & Schielzeth, H. (2017). The coefficient of determination R^2 and intra-class correlation coefficient from generalized linear mixed-effects models revisited and expanded. *Journal of the Royal Society Interface*, *14*, 20170213.

Nevo, E., Fu, Y. B., Pavlicek, T., Khalifa, S., Tavasi, M., & Beiles, A. (2012). Evolution of wild cereals during 28 years of global warming in Israel. *Proceedings of the National Academy of Sciences of the United States of America*, *109*, 3412–3415.

Paccard, A., Vance, M., & Willi, Y. (2013). Weak impact of fine-scale landscape heterogeneity on evolutionary potential in *Arabidopsis lyrata*. *Journal of Evolutionary Biology*, *26*, 2331–2340.

Peakall, R., & Smouse, P. E. (2012). GenAlEx 6.5: Genetic analysis in Excel. Population genetic software for teaching and research-an update. *Bioinformatics*, *28*, 2537–2539.

Picó, F. X. (2012). Demographic fate of *Arabidopsis thaliana* cohorts of autumn- and spring-germinated plants along an altitudinal gradient. *Journal of Ecology*, *100*, 1009–1018.

Picó, F. X. (2022). Data from: Spatio-temporal dynamics of genetic variation at the quantitative and molecular levels within a natural *Arabidopsis thaliana* population. *Dryad Digital Repository*, https://doi.org/10.5061/dryad.7sqv9s4vt

Picó, F. X., Méndez-Vigo, B., Martínez-Zapater, J. M., & Alonso-Blanco, C. (2008). Natural genetic variation of *Arabidopsis thaliana* is geographically structured in the Iberian Peninsula. *Genetics*, *180*, 1009–1021.

Postma, F. M., & Ågren, J. (2016). Early life stages contribute strongly to local adaptation in *Arabidopsis thaliana*. *Proceedings of the National Academy of Sciences of the United States of America*, *113*, 7590–7595.

Postma, F. M., Lundemo, S., & Ågren, J. (2016). Seed dormancy cycling and mortality differ between two locally adapted populations of *Arabidopsis thaliana*. *Annals of Botany*, *117*, 249–256.

Prati, D., & Schmid, B. (2000). Genetic differentiation of life-history traits within populations of the clonal plant *Ranunculus reptans*. *Oikos*, *90*, 442–456.

Quintana-Ascencio, P. F., Koontz, S. M., Smith, S. A., Sclater, V. L., David, A. S., & Menges, E. S. (2018). Predicting landscape-level distribution and abundance: Integrating demography, fire, elevation and landscape habitat configuration. *Journal of Ecology*, *106*, 2395–2408.

Rangel, T. F., Diniz-Filho, J. A. F., & Bini, L. M. (2010). SAM: A comprehensive application for spatial analysis in macroecology. *Ecography*, *33*, 46–50.

Rosenberg, M. S., & Anderson, C. D. (2011). PASSaGE: Pattern analysis, spatial statistics and geographic exegesis. Version 2. *Methods in Ecololy and Evolution*, *2*, 229–232.

Sánchez-Bermejo, E., Méndez-Vigo, B., Picó, F. X., Martínez-Zapater, J. M., & Alonso-Blanco, C. (2012). Novel natural alleles at *FLC* and *LVR* loci account for enhanced vernalization responses in *Arabidopsis thaliana*. *Plant, Cell and Environment*, *35*, 1672–1684.

Scarcelli, N., Cheverud, J. M., Schaal, B. A., & Kover, P. X. (2007). Antagonistic pleiotropic effects reduce the potential adaptive value of the *FRIGIDA* locus. *Proceedings of the National Academy of Sciences of the United States of America*, *104*, 16986–16991.

Schemske, D. W. (1984). Population structure and local selection in *Impatiens pallida* (Balsaminaceae), a selfing annual. *Evolution*, *38*, 817–832.

Siepielski, A. M., DiBattista, J. D., & Carlson, S. M. (2009). It's about time: The temporal dynamics of phenotypic selection in the wild. *Ecology Letters*, *12*, 1261–1276.

Stenøien, H. K., Fenster, C. B., Tonteri, A., & Savolainen, O. (2005). Genetic variability in natural populations of *Arabidopsis thaliana* in northern Europe. *Molecular Ecology*, *14*, 137–148.

Stratton, D. A., & Bennington, C. C. (1996). Measuring spatial variation in natural selection using randomly-sown seeds of *Arabidopsis thaliana*. *Journal of Evolutionary Biology*, *9*, 215–228.

Tabas-Madrid, D., Méndez-Vigo, B., Arteaga, N., Marcer, A., Pascual-Montano, A., Weigel, D., Xavier Picó, F., & Alonso-Blanco, C. (2018). Genome-wide signatures of flowering adaptation to climate temperature: Regional analyses in a highly diverse native range of *Arabidopsis thaliana*. *Plant, Cell and Environment*, *41*, 1806–1820.

Takou, M., Wieters, B., Kopriva, S., Coupland, G., Linstädter, A., & de Meaux, J. (2019). Linking genes with ecological strategies in *Arabidopsis thaliana*. *Journal of Experimental Botany*, *70*, 1141–1151.

Taylor, M. A., Cooper, M. D., Sellamuthu, R., Braun, P., Migneault, A., Browning, A., Perry, E., & Schmitt, J. (2017). Interacting effects of genetic variation for seed dormancy and flowering time on phenology, life history, and fitness of experimental *Arabidopsis thaliana* populations over multiple generations in the field. *New Phytologist*, *216*, 291–302.

The 1001 Genomes Consortium. (2016). 1,135 Genomes reveal the global pattern of polymorphism in *Arabidopsis thaliana*. *Cell*, *166*, 481–491.

Toledo, B., Marcer, A., Méndez-Vigo, B., Alonso-Blanco, C., & Picó, F. X. (2020). An ecological history of the relict genetic lineage of *Arabidopsis thaliana*. *Environmental and Experimental Botany*, *170*, 103800.

Vekemans, X., & Hardy, O. J. (2004). New insights from fine-scale spatial genetic structure analyses in plant populations. *Molecular Ecology*, *13*, 921–935.

Vidigal, D. S., Marques, A. C. S. S., Willems, L. A. J., Buijs, G., Méndez-Vigo, B., Hilhorst, H. W. M., Bentsink, L., Picó, F. X., & Alonso-Blanco, C. (2016). Altitudinal and climatic associations of seed dormancy and flowering traits evidence adaptation of annual life cycle timing in *Arabidopsis thaliana*. *Plant, Cell and Environment*, *39*, 1737–1748.

Volis, S., Ormanbekova, D., & Shulgina, I. (2016). Fine-scale spatial genetic structure in predominantly selfing plants with limited seed dispersal: A rule or exception? *Plant Diversity*, *38*, 59–64.

Weinig, C., Dorn, L. A., Kane, N. C., German, Z. M., Halldorsdottir, S. S., Ungerer, M. C., Toyonaga, Y., Mackay, T. F. C., Purugganan, M. D., & Schmitt, J. (2003). Heterogeneous selection at specific loci in natural environments in *Arabidopsis thaliana*. *Genetics*, *165*, 321–329.

Wiegand, T., & Moloney, A. K. (2004). Rings, circles, and null-models for point pattern analysis in ecology. *Oikos*, *104*, 209–229.

Wilczek, A. M., Cooper, M. D., Korves, T. M., & Schmitt, J. (2014). Lagging adaptation to warming climate in *Arabidopsis thaliana*. *Proceedings of the National Academy of Sciences of the United States of America*, *111*, 7906–7913.

Wilczek, A. M., Roe, J. L., Knapp, M. C., Cooper, M. D., Lopez-Gallego, C., Martin, L. J., Muir, C. D., Sim, S., Walker, A., Anderson, J., Egan, J. F., Moyers, B. T., Petipas, R., Giakountis, A., Charbit, E., Coupland, G., Welch, S. M., & Schmitt, J. (2009). Effects of genetic perturbation on seasonal life history plasticity. *Science*, *323*, 930–934.

Wittmann, M. J., Bergland, A. O., Feldman, M. W., Schmidt, P. S., & Petrov, D. A. (2017). Seasonally fluctuating selection can maintain polymorphism at many loci via segregation lift. *Proceedings of the National Academy of Sciences of the United States of America*, *114*, E9932–E9941.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Méndez-Vigo, B., Castilla, A. R., Gómez, R., Marcer, A., Alonso-Blanco, C., & Picó, F. X. (2022). Spatiotemporal dynamics of genetic variation at the quantitative and molecular levels within a natural *Arabidopsis thaliana* population. *Journal of Ecology*, *110*, 2701–2716. https://doi.org/10.1111/1365-2745.13981