Adaptation and extinction in experimentally fragmented landscapes

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Competition and disturbance are potent ecological forces that shape evolutionary trajectories. These forces typically work in opposition: when disturbance is infrequent, densities are high and competition is intense. In contrast, frequent disturbance creates a low-density environment in which competition is weak and good dispersal essential. We exploited recent advances in genomic research to quantify the response to selection by these powerful ecological forces at the phenotypic and molecular genetic level in experimental landscapes. We grew the annual plant *Arabidopsis thaliana* in discrete patches embedded in a hostile matrix and varied the number and size of patches and the intensity of disturbance, by creating both static and dynamic landscapes. In static landscapes all patches were undisturbed, whereas in dynamic landscapes all patches were destroyed in each generation, forcing seeds to disperse to new locations. We measured the resulting changes in phenotypic, genetic, and genotypic diversity after five generations of selection. Simulations revealed that the observed loss of genetic diversity dwarfed that expected under drift, with dramatic diversity loss, particularly from dynamic landscapes. In line with ecological theory, static landscapes favored good competitors; however, competitive ability was linked to growth rate and not, as expected, to seed mass. In dynamic landscapes, there was strong selection for increased dispersal ability in the form of increased inflorescence height and reduced seed mass. The most competitive genotypes were almost eliminated from highly disturbed landscapes, raising concern over the impact of increased levels of human-induced disturbance in natural landscapes.

Arabidopsis thaliana | competition | disturbance | evolution | genomics

Natural selection should lead to adaptive evolution in response to ecological forces. However, in spatially structured landscapes, competition and disturbance can exert conflicting selection pressures. For example, high disturbance rates lead to reduced densities and reduced competition but favor those traits that confer good dispersal ability. In contrast, low disturbance rates lead to high density and intense competition, favoring traits that confer good competitive ability (1–3). However, although competition and disturbance both lead to exclusion and loss of diversity, coexistence can theoretically occur in multispecies communities at some intermediate disturbance level (4–7).

Although the conditions for coexistence have been a subject of great debate (7), the relative strengths of disturbance and competition as forces shaping the phenotypic and genetic composition of plant communities have rarely been examined in an experimental setting. Here we used communities composed of multiple genotypes of *Arabidopsis thaliana* (L.) Heynh. to address this question. We considered this to be an appropriate model for an ecological community because natural populations of *Arabidopsis thaliana* are >95% self-fertilizing (8); hence, like a group of species, cooccurring lines mostly produce seeds of a genotype identical to the parent. In addition, recombinant inbred line (RIL) populations are available for *Arabidopsis* (9); these populations have no coevolutionary history, so that the success of genotypes can be more easily linked to particular genes and traits. Moreover, changes in the frequency of alleles can be measured using high-throughput genomic methods, localized to specific regions of the chromosomes and related to known quantitative trait loci (QTLs) for the relevant traits (10, 11).

We constructed 24 independent experimental landscapes in a glass house, each consisting of multiple habitat patches embedded in a hostile matrix. Because our primary focus was not coexistence, we imposed two disturbance regimes (static and dynamic) representing the extreme ends of the disturbance gradient: in static landscapes patches were never disturbed, whereas in dynamic landscapes all patches were destroyed every generation and only dispersing seeds survived. To create static landscapes, nondispersing seeds were collected from the surface of existing patches, and all dispersing seeds falling into the matrix were destroyed (Fig. 1*A*). To create dynamic landscapes we collected dispersing seeds by placing randomly arranged Petri dishes of the same size and number as the original patches within the matrix and destroyed all existing patches at the end of each generation (Fig. 1*A*). In all landscapes, patches were then randomly relocated, refilled with new soil, and sown with seeds collected from the previous generation. Landscapes consisted of 2, 4, 8, or 16 patches, with the combined patch area held constant at approximately 7% of the total [similar to the percentage of bare earth available in sand dunes where *Arabidopsis* naturally occurs (12)]. We included a patch number treatment because increasing the number of patches decreases the average dispersal distance and allows a greater heterogeneity to develop among patches, potentially slowing competitive exclusion.

We seeded the landscapes with a selection of RILs derived from the large-seeded Cape Verde Islands (Cvi) and the small-seeded Landsberg *erecta* (L*er*) accessions (13, 14). We chose this population because it exhibits a seed size/number tradeoff (15) and because competition/colonization tradeoffs have often been cast in terms of seed size (16, 17): large-seeded species are suggested to have superior competitive ability and small-seeded

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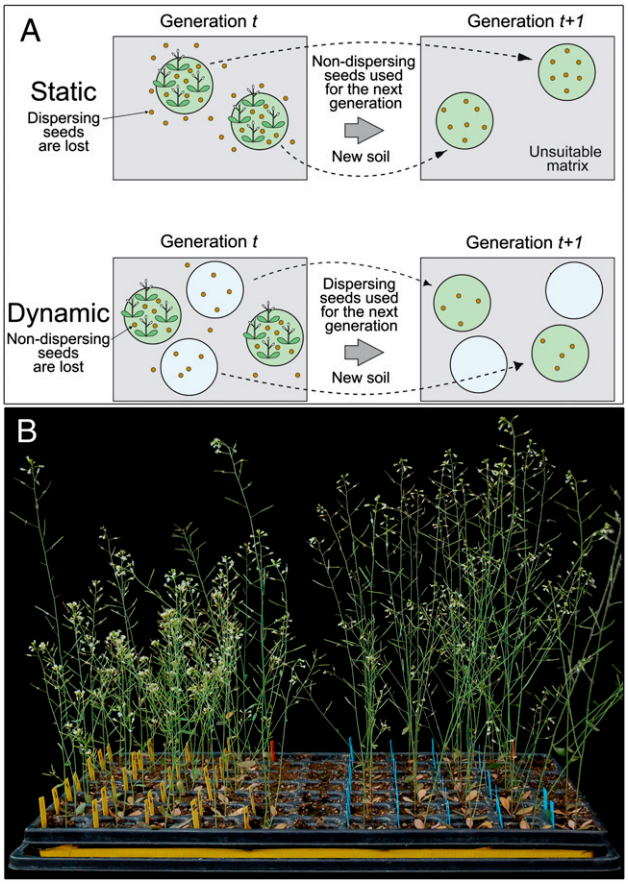

Fig. 1. Comparison of plants sampled from static and dynamic landscapes after five generations of selection. (*A*) Each experimental landscape was initially sown with seeds in generation 1. Natural seed production then provided new seeds for all subsequent generations. In static landscapes, seeds were collected from the surface of existing patches, whereas in dynamic landscapes, randomly arranged Petri dishes of the same size and number as the original patches were set out in the landscapes to collect dispersing seeds. (*B*) Plants grown under standardized conditions (one plant per cell) from seeds sampled from generation-5 plants of the selection experiment. Individuals from a representative static (*Left*) and a representative dynamic (*Right*) landscape are shown.

species superior colonizing abilities (because they produce more seeds). In addition, because inflorescence height will inevitably be linked to dispersal distances, we exploited the presence of the *erecta* mutation in this population, which greatly reduces inflorescence height. Of the 19 lines selected, 10 carried the *erecta* mutation and 9 carried the wild-type *ERECTA* allele (*SI Appendix*, Table S1). When grown under our conditions the mean mass per 100 seeds ranged from 2.34 mg to 5.07 mg among lines. Among the selected lines, seed mass was unaffected by the *erecta* mutation ($t = 0.156$, df = 17, $P = 0.8$), whereas inflorescence height was considerably reduced ($t = 5.94$, df = 17, $P < 0.0001$); thus, selection can act mostly independently on inflorescence height and seed mass. After seeding the 24 landscapes in generation 1, they were allowed to evolve independently for five generations with no mixing among landscapes.

Results

Population Density. Seedling and adult densities were recorded in all landscapes in each generation. In static landscapes, mean seedling densities were higher [static: 700.7 (95% CI, 614–787); dynamic: 341.5 (95% CI, 255–428)], as expected if most seeds fall close to the parent plants, and a much smaller fraction of those seedlings survived to adulthood as compared with dynamic landscapes ($F_{1,20} = 76.99$, $P < 0.0001$). To test whether the reduced survival of seedlings in static landscapes is due to higher seedling densities, we refitted the model with seedling density fitted first as covariate. Seedling density had a highly significant negative effect on survival ($F_{1,208} = 557.8$, $P < 0.0001$), and fitting this covariate removed the significant difference between static and dynamic landscapes ($F_{1,20} = 1.43$, $P = 0.25$), supporting the idea that the reduced survival observed in static landscapes is caused by increased competition. Seedlings also survived better in landscapes with larger patch sizes ($F_{1,20} = 73.67$, $P < 0.0001$), but there was no effect of patch size on seedling densities ($F_{1,20} = 0.53$, $P = 0.47$; *SI Appendix*, Table S2).

Phenotypic Changes. Measurements of plant height and seed mass in each generation indicated that static and dynamic populations gradually diverged through time (*SI Appendix*, Fig. S1), although there were no clear or consistent effects of patch size. To remove the confounding effects of density, seeds were sampled from individuals growing in all 24 landscapes in generation 5 and sown in single cells. To control for maternal effects, seeds from these individuals were then collected and resown in single cells alongside four individuals from each of the 19 lines forming the original, ancestral population. After growing in standardized conditions, plants sampled from dynamic landscapes were clearly taller than those from static landscapes, a difference that was due to two separate phenomena (Figs. 1*B* and 2). First, the percentage of individuals carrying the *erecta* mutation was much lower in dynamic landscapes compared with static ones [static: 44.1% (95% CI, 40.8–47.5%) *erecta*; dynamic: 7.81% (95% CI, 6.04–9.85%) *erecta*]. Second, both mutant and wild-type individuals were taller in dynamic landscapes: individuals carrying the *erecta* mutation were on average 2.5 (95% CI, 0.088–5.9) cm taller, whereas wild-type *ERECTA* individuals were on average 9.0 (95% CI, 7.4–10.6) cm taller (*SI Appendix*, Table S2).

Contrary to our expectation that competitive environments would favor large seeds, the average seed mass in static landscapes [mass of 100 seeds: 2.65 (95% CI, 2.14–3.16) mg] was not significantly different ($F_{1,20} = 2.98$, $P = 0.099$) from that in dynamic ones [mass of 100 seeds: 2.39 (95% CI, 2.18–2.61) mg]. However, populations in all landscapes experienced selection for lighter seeds compared with the ancestral population [mass of 100 seeds: 3.32 (95% CI, 3.10–3.55) mg; Fig. 2]. Nevertheless, populations from static landscapes had significantly higher variance in seed mass ($F_{1,22} = 14.5$, $P < 0.001$), owing to the higher frequency of large-seeded individuals: for example, the percentage of individuals with seed mass greater than the ancestral mean was 22.4% in static landscapes but only 4.1% in dynamic landscapes. It therefore seems that much stronger directional selection occurred in dynamic landscapes, leading to greater loss of phenotypic diversity.

Genome-Wide Genetic Changes. To characterize the observed phenotypic changes at the genotypic level we carried out a genome-wide genetic analysis of the 24 populations using rapid array mapping (10). In a RIL population a maximum of two different alleles are present at any given locus (18) because all lines are derived from only two homozygous parental accessions (in our case L*er* and Cvi); hence, we can examine changes in the frequency of L*er* and Cvi alleles across the genome in populations after selection. First, we used an efficient genotyping approach using ATH1 Affymetrix Genechips to identify genes that harbor two different alleles in the parental accessions (ArrayExpress, accession no. E-MTAB-107)—so-called single feature polymorphisms (SFPs) (19). SFPs could be any genetic mismatch (e.g., deletion, nucleotide changes) that results in significant differences in hybridization intensities between the two parental lines on the microarrays. Comparing the normalized and averaged microarray

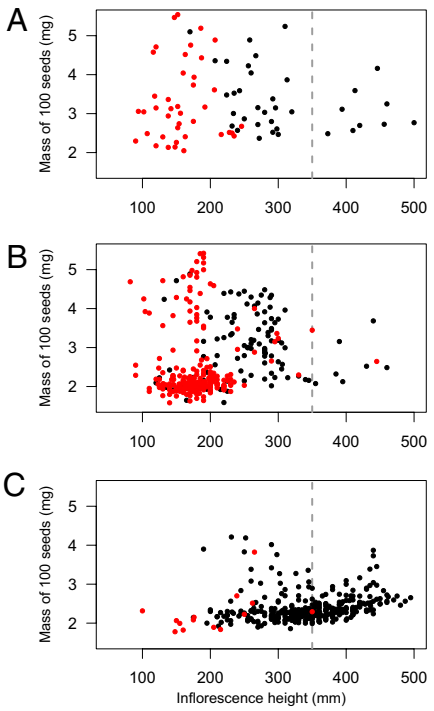

Fig. 2. Comparison of seed mass and inflorescence height in the ancestral population and in populations after five generations of selection. (*A*) Inflorescence height and seed mass of individuals in the ancestral population (measured on four different individuals from each of the 19 lines). (*B*) The same traits measured on 30 different individuals from each of the 12 static landscapes; and (*C*) the same traits measured on 30 different individuals from each of the 12 dynamic landscapes. Plants carrying the *erecta* mutation are shown in red. Plants with height >350 mm are highlighted by the gray dotted line. Among the ancestral population, there are nine such individuals belonging to only three RILs.

signal ratios between the two parents (L*er* and Cvi), we mapped 26,638 SFPs at a false discovery rate of 4.8%, allowing us to unambiguously distinguish the L*er* and Cvi alleles at more than 26,000 loci. This provided a very dense physical map with 213 molecular markers per megabase across the *Arabidopsis* genome. For each of the SFP markers, bulk segregant analysis (10) was carried out using microarrays to calculate allele frequencies for each of the 24 populations after selection relative to the ancestral population.

To have roughly equal contributions of genetic material from all individuals within each population, we picked one flower per progeny of each generation-5 plant grown under standardized conditions. Within each of the 24 populations, flowers were pooled for DNA extraction and microarray hybridization. For each population, the normalized data were first scaled according to the differences in mean hybridization intensities of the parents. Scaled signals from the ancestral population (17 RILs plus two parental lines) were subtracted to correct for any bias in allele frequencies in the ancestral population, and the data were LOESS smoothed. If there was no change in allele frequencies, ratios should center on zero (heterozygous), whereas if Cvi alleles were preferentially selected, ratios should move toward +0.5 (homozygous Cvi), and if L*er* alleles were preferentially selected, ratios should move toward −0.5 (homozygous L*er*).

Generally, allele frequencies in populations from both static and dynamic landscapes moved toward L*er* (Fig. 3); however, the average shift in dynamic landscapes was significantly greater than in static ones, and populations from static landscapes were significantly more heterozygous, again suggesting that directional selection has been less intense (*SI Appendix*, Fig. S2). We compared our profiles with the positions of previously identified QTLs (14, 20–22), which are found across the *Arabidopsis* genome and affect many phenotypic traits, including seed mass and plant height (Fig. 3 and *SI Appendix*, Fig. S3). Six major QTLs (Fig. 3, green rectangles) have been identified in *Arabidopsis*, which together influence an estimated 77% of all phenotypic traits analyzed to date (20). All but one of these major QTLs map to regions that show significant allele frequency shifts toward L*er* in populations from dynamic landscapes but remain heterozygous in populations from static landscapes. The major QTL on chromosome 2 maps to the *erecta* mutation, an important regulator of plant height, and is the only part of the genome that has significantly shifted toward Cvi in dynamic landscapes. The frequency of the *erecta* mutation in the ancestral population was 52.6%. In populations from dynamic landscapes the genetic estimate of the mean frequency of *erecta* plants was 15.61% (95% CI, 9.96–21.26%), compared with 46.12% (95% CI, 36.5–55.74%) in populations from static landscapes, in good agreement with the phenotypic data. It therefore seems that the *erecta* mutation was selectively neutral in static landscapes but experienced strong, negative selection in dynamic ones. Allele frequencies along the entire length of chromosome 4 have shifted toward L*er* in both landscape types. Previously mapped QTLs for plant height and seed mass on chromosome 4 are not focused around a major QTL (in contrast to chromosomes 1, 2, 3, and 5, where they are mostly found to colocalize with major QTLs) but are instead distributed across the entire chromosome (*SI Appendix*, Fig. S3).

Genotypic Changes. Each RIL has a unique pattern of L*er* and Cvi alleles (9), yielding a distinctive chromosomal signature and allowing us to identify successful lines. We genotyped individuals from both 16-patch static (n = 120) and 16-patch dynamic (n = 118) landscapes, which revealed that lines generally bred true, although four recombinant individuals were found. To assess the possible magnitude of drift, we simulated genotypic diversity in generation 5 assuming that, in each generation, genotypes were selected at random according only to their frequency in the previous generation. Population sizes were constrained to the known adult population sizes. We repeated this 5,000 times for each landscape to construct a confidence interval for the expected genotypic diversity under drift alone (*SI Appendix*, Fig. S4). The simulations reveal that the observed diversity loss in both static and dynamic landscapes far exceeds that expected under drift alone (*SI Appendix*, Fig. S4).

In dynamic landscapes there was strong selection for genotypes producing a tall inflorescence; for example, 43.1% of all individuals in dynamic landscapes had inflorescence height >350 mm, and such plants in the ancestral population (Fig. 2) belong to only three RILs. In generation 5, two of these three lines (CVL39 and CVL125) made up 90% of all genotyped individuals (Fig. 4). Both lines have very small seeds and consist predominantly of L*er* alleles: CVL125 carries L*er* alleles at an estimated 90% and CVL39 at 77% of the genome; hence they are genetically identical at 78% of the genome.

In populations from static landscapes, 10 of the original 19 lines were found (Fig. 4), although, unlike in dynamic landscapes, these genotypes vary considerably in both seed mass and inflorescence height. In a separate experiment, we therefore measured size-standardized growth rates on the 17 RILs plus the L*er* parent (23). Growth rate was a very good predictor of abundance in static landscapes ($F_{1,13}$ = 14.56, P = 0.0019), unlike inflorescence height ($F_{1,13}$ = 1.14, P = 0.31), the presence of the *erecta* mutation ($F_{1,13}$ = 0.73, P = 0.41), or seed mass ($F_{1,13}$ = 1.71, P = 0.21). Rapid growth is likely to be selected when there is intense scramble competition for resources, as is likely to occur among synchronously germinating annual plants (24, 25). The two most successful lines in static landscapes were the small-seeded

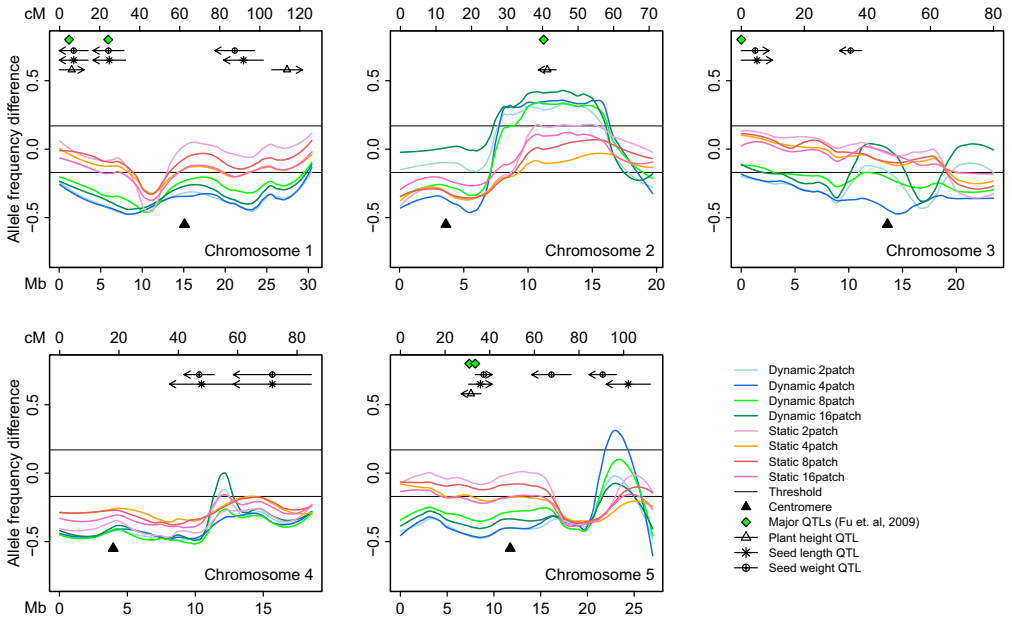

Fig. 3. Allele frequency shifts along chromosomes for dynamic and static landscapes. Features were plotted on the physical map (Mb); the genetic Map (cM) is given as reference. The mean of the three replicate populations within each of the eight treatment combinations is shown. The threshold for significant frequency shifts toward Cvi alleles is +0.17, and the threshold for shifts toward L*er* alleles is −0.17 (11, 33). The position of previously identified major QTLs (20) and QTLs for seed weight, seed length, and plant height (13), as well as centromeres, are indicated. Arrows mark the 2-Logarithm of Odds (2-LOD) support interval of QTLs. Left-pointing arrows indicate that the Cvi allele increases the phenotypic value of the trait, right-pointing arrows that the Cvi allele decreases the phenotypic value of the trait. Information on the variance in a QTL affecting a particular phenotypic trait has been published previously (22) and is not included in the figure.

L*er* parent and the large-seeded CVL168 (Fig. 4). These two fast-growing genotypes made up 49% of individuals in static landscapes, but they were almost eliminated from dynamic landscapes, presumably because they are both short and hence have poor dispersal ability.

Discussion

After only five generations, populations in static and dynamic landscapes had diverged both phenotypically and genetically and were dominated by different genotypes. This supports the basic assumption of competition/colonization tradeoff models, that competition and disturbance select for fundamentally different traits, and that a single species (or genotype) is unlikely to be both a good competitor and a good colonizer. However, although both competition and disturbance exerted directional selection, leading to exclusion and loss of diversity, disturbance was the more potent force in this regard. In contrast, the effects of patch size were weak and nonsignificant, perhaps because the limited scale of our experiment precluded such important ecological phenomena as edge effects, in which individuals growing near the edge of patches are particularly disadvantaged.

Competition/colonization tradeoff models usually seek to explain the coexistence of species with different traits, which we did not assess here; for example, if large seeds have increased competitive ability (16, 17), then a competition/colonization tradeoff model can potentially support multiple seed size strategies. However, the mean seed mass in static landscapes declined relative to the ancestral population, making this an unlikely explanation (26). Instead, success in static landscapes was strongly correlated with growth rate. In highly disturbed or dynamic landscapes, two very tall, small-seeded genotypes almost exclusively dominated. Inflorescence height was clearly the most important trait for success in dynamic landscapes, because small-seeded genotypes with short inflorescences (those carrying the *erecta* mutation) were not successful. Such rapid evolution for good dispersal ability probably occurs because a small difference in mean height can greatly increase the chances of a seed dispersing a relatively long distance (27, 28); hence, short genotypes were heavily penalized in dynamic landscapes. It is noteworthy that the sensitivity of highly competitive genotypes to disturbance and their loss from dynamic landscapes is also predicted by competition/colonization tradeoff models (29). Thus, the increasing levels of human-induced disturbance in natural habitats can be expected to have phenotypic and genetic consequences.

Recent theoretical developments in community ecology have suggested alternative models of community diversity (30) that rely on equalizing tradeoffs (31)—to reduce fitness differences among species with different traits—coupled with ecological drift. However, despite small population sizes, selection was still the dominant force in our experiment. This demonstrates that traits closely linked to fitness, such as height, seed size, and growth rate, are unlikely to be selectively neutral, even in the presence of tradeoffs that tend to equalize fitness differences (32). Our results also demonstrate that although certain traits or genes may be close to neutral in one particular environment (e.g., the *erecta* mutation in static landscapes), it is highly unlikely that this neutrality will be preserved under any major environmental change. Thus, the extreme fragility of neutral models makes them unlikely candidates for the long-term maintenance of trait diversity.

Materials and Methods

Landscapes. Landscapes were trays measuring 90 × 64 cm and 7 cm deep, filled with a sand/soil mixture. Landscapes contained 2, 4, 8, or 16 circular patches (cylindrical slices of PVC tubing cut to the same depth as the tray) with diameters of 17.5 cm, 11 cm, 8 cm, or 5.7 cm, respectively, thus keeping the total suitable area roughly constant at approximately 7%. Landscapes were set up in a glass house and subjected to two levels of disturbance (static or dynamic). Patches were located in a stratified random way and relocated in each generation. Each landscape was sown with 16 seeds of each of the 19 lines in generation 1, but in subsequent generations dispersing seeds (dynamic landscapes) and nondispersing seeds (static landscapes) were collected and transferred to the next generation (Fig. 1). Each landscape was

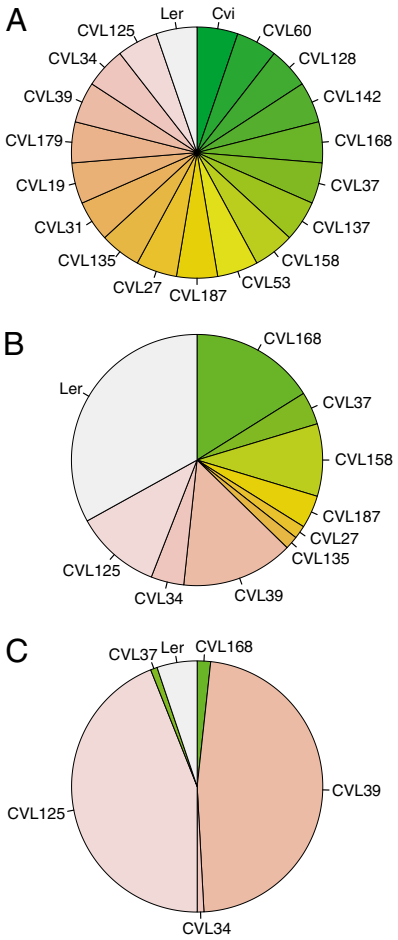

Fig. 4. Frequencies of the 19 lines before and after five generations of selection. (*A*) In the ancestral population, 19 lines were sown at equal frequency. Colors reflect the percentage of alleles in each RIL inherited from the Landsberg (L*er*) parent (pink = high percentage L*er* alleles; green = low percentage L*er* alleles). In static (*B*) and dynamic (*C*) landscapes, a sample of individuals from the 16-patch landscapes (static: $n = 120$; dynamic: $n = 118$) were genotyped after five generations of selection.

surrounded by fine netting to prevent seed dispersal among landscapes. Landscapes were irrigated by flooding the table twice per day. Nine weeks after sowing, all plants were removed and half the plants were measured. We kept a minimum daytime temperature of 22 °C and a minimum night temperature of 20 °C. Additional lighting came on automatically when daylight fell below 25 klx and hence we ensured a 16-h day in all generations.

RILs. The RIL population is described fully elsewhere (13, 14). We selected lines for this experiment by sampling across the entire available seed mass range with a balance of mutant *erecta* and wild-type *ERECTA* lines.

Phenotypic Assessment. In generation 5, a single silique was removed from 77 randomly selected plants in each landscape. Seeds from different siliques were not mixed, and we regrew the 77 individuals from each landscape under standardized conditions (isolated cells in flats filled with the same sand/soil mixture used in the main experiment) to exclude the effects of density on the phenotype. The presence of the *erecta* mutation was scored by eye by independent experts (Masaki Kobayahi and Matthias Helling, Institute of Plant Biology, University of Zürich). A single flower was also removed from each of these plants for the genetic analysis described below (54–77 individuals per landscape). To eliminate potential maternal effects, we resampled seeds from 30 of the 77 individuals from each landscape growing under standardized conditions and grew these 30 individuals a second time in the same way. The 30 individuals were chosen at random from the pool of 77. This time we measured on each individual: germination day, plant height, and the total number of seed pods. In addition, two seed pods were removed from each plant (the lowest seed pods on the main stem), and all seeds within these two pods were counted and weighed. This enabled us to determine the total mass of seeds produced and the average seed mass of the individual.

Statistical analysis of phenotypic traits was carried out using linear mixed-effect models in the R statistical package (R Foundation for Statistical Computing) with landscape number (1–24) as the random effect. Patch area was log-transformed and fitted as a continuous variable. For measurements made in each generation (seedling counts and survival), Generation (1–5) was fitted as a factor. To analyze seedling survival, the fraction Adults/Seedlings was log-transformed because in ≈20% of cases the number of adults was actually greater than the number of seedlings, presumably because of late seedling emergence. More details of statistical analyses are available in *SI Appendix*, Table S2.

Genotypic Assessment. Bulk segregant mapping with array genotyping has been described in detail elsewhere (11). Analysis scripts were adapted as follows (available upon request): Because the parental accessions were L*er* and Cvi and not Columbia (Col), which is the basis of the targets located on the ATH1 Affymetrix Genechips, two-tailed modified *t* tests were performed instead of one-tailed. To minimize the possibility that identified SFPs represent duplications between Cvi and L*er* accessions, potential SFP duplications were mapped using a two-tailed *t* test on previously published microarray data for Col/L*er* and Col/Cvi. Hybridization signals that were significantly higher in the L*er* or Cvi accessions compared with the Col accession indicate duplications. A threshold of 15,011 (Col/L*er*) and 15,133 (Col/Cvi) SFP hits was used to identify 1,382 and 754 potential duplications, respectively (the total number of SFP hits for Cvi/L*er* was 27,692, which was considered the minimum combined threshold). Of these 2,136 potential duplications, 1,937 were unique and 1,054 SFP markers matched in the Cvi/L*er* population and were thus removed from further analysis, leaving a total of 26,638 SFP markers for bulk segregant analysis. Raw hybridization data for mapping duplications are available upon request. Simulation studies have been used to set appropriate thresholds for significant allele frequency shifts for different genetic models and pool sizes (11, 33). We used a threshold of ± 0.17, which is appropriate for 60 plants per pool (landscape scenario). R scripts were used for figure plotting. Raw data and normalized data from microarray experiments have been published in ArrayExpress (accession no. E-MTAB-107).

Mapping of Genetic Markers. Because QTL data were published using genetic markers (cM), and the microarray data were generated using the published *Arabidopsis* sequence (AGI, 125 Mb), we mapped the genetic markers onto the nucleotide sequence. Two different genetic maps were used for mapping QTLs and genotyping RIL lines. For QTL mapping using Col/L*er* RIL lines (21, 22) we used the NASC genetic Map (http://arabidopsis.info/new_ri_map.html) with a total of 1,288 genetic markers. Markers with known primer sequences were mapped to the genome sequence using BLAST analysis. All remaining markers for known nucleotide positions were identified by searching the TAIR database (http://www.arabidopsis.org/index.jsp). In total, 590 genetic markers were mapped to their physical position, which corresponds to a density of 1/212 kb. Physical markers that mapped on or close to the published QTL positions were used for mapping QTLs onto the genome sequence. For QTL mapping of Cvi/L*er*-derived RILs and Cvi/L*er* RIL genotyping we used the published AFLP Cvi/L*er* RIL map (9). This map consists of 292 genetic markers, of which 115 were previously mapped to unique nucleotide positions in the genome (34). For the remaining markers not previously mapped, the nucleotide position was estimated by interpolation with the position of the closest marker for which the nucleotide position was known. Using this information, markers used in RIL genotyping as well as QTL markers were mapped to their respective positions in the genome sequence. We noticed a shift of exactly 1 Mb on chromosome 4 in the originally published physical AFLP map (34) compared with the published AGI sequence and corrected marker positions accordingly. All data in the figures were plotted on the genome sequence scale (Mb); the genetic scale (cM) is given as reference only.

Genotyping Lines. We sampled seeds from 40 different generation-5 plants in each of the 16-patch landscapes (three static and three dynamic). Seeds were plated out on MS-agar, and 120 seedlings per landscape type were grown for 14 d before DNA was extracted (35) (96-well format) for genotyping. To distinguish the 17 RIL lines and the two parental lines, we tested nine polymorphic genomic loci (36) that gave different fragment size on amplification (fragment length polymorphism) in the L*er* and the Cvi parental

lines using PCR. A summary of the markers used, the PCR primers, and the resulting fragment length polymorphism is provided in *SI Appendix*, Table S3. PCR was first carried out on DNA of the 17 ancestral RIL lines to establish the reference genotype. The PCR conditions were as follows: 94 °C for 120 s (1×); followed by 35 cycles of 94 °C for 30 s, 60 °C for 30 s (primer 9 for 90 s), 72 °C for 45 s; and 72 °C for 10 min (1×). The size of PCR products was analyzed on a 2% agarose gel.

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Supporting Information

Figure S1. The divergence of plant height and seed mass observed through time in static vs. dynamic landscapes. (**a**) Normalised plant height scores calculated for each landscape based on measurements of half the adult plants in generations 1–5. (**b**) Normalised seed mass scores for each landscape based on three bulk samples each consisting of 16 seeds taken from 16 different individuals in generations 2–5. The Z-score for each landscape is calculated using the mean and standard deviation of each trait in each generation. The mean and 95% confidence interval are shown.

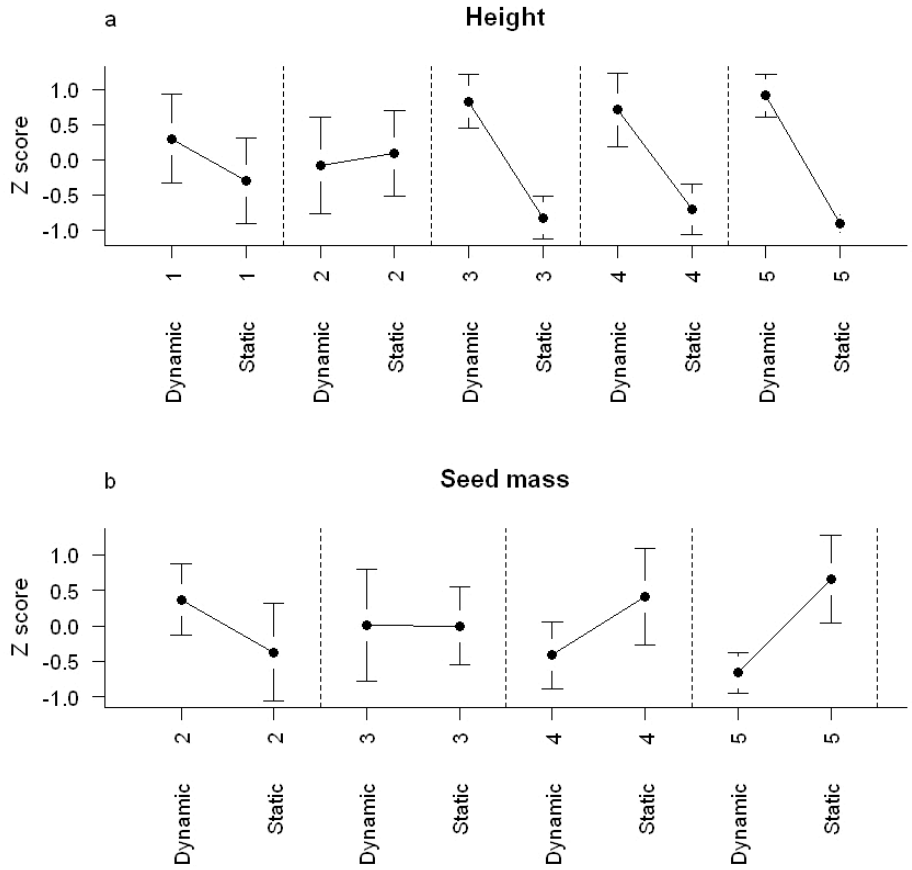

Figure S2. Analysis of average allele frequencies and homozygosity in dynamic and static landscapes. (**a**) the mean allele frequencies for SFP markers along all chromosomes. The data from each landscape were LOESS smoothed after subtraction of the original signal. In both static and dynamic landscapes there has been a shift towards L*er* (i.e. towards –0.5), but this shift is stronger in dynamic landscapes. (**b**) The degree to which SFP markers are homozygous, calculated by removing the sign of the absolute score and then averaging. The populations in dynamic landscapes are more homozygous compared to static ones, although the confidence interval does not include zero in either case indicating that populations are more homozygous after selection in both landscape types. The mean and 95% confidence interval are shown.

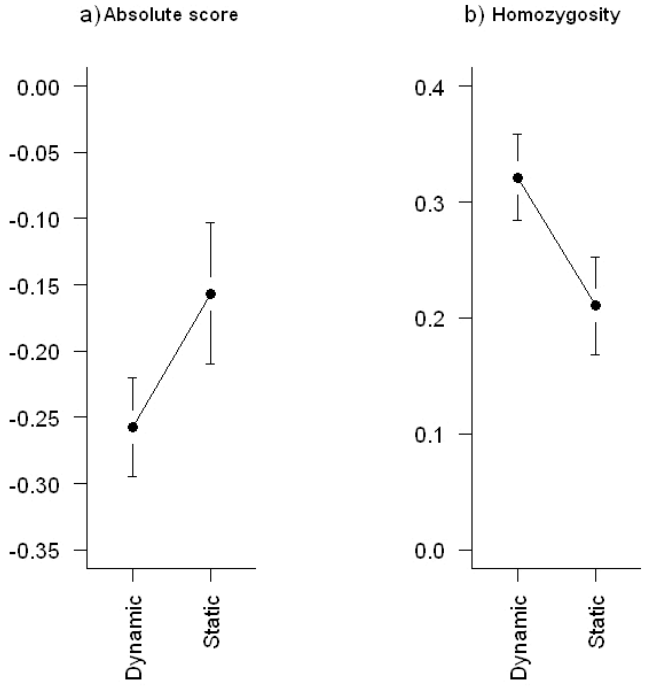

Figure S3. Previously identified Quantitative Trait Loci (QTLs) co-localise with allele frequency shifts in dynamic and static landscapes. QTLs identified in four different studies are plotted along with the allele frequency patterns. QTLs for each trait are depicted with different symbols and arrows or horizontal lines associated with each QTL mark the 2-LOD support intervals. (**a**) QTLs mapped in a study investigating seed size loci in relation to other life history traits using the Cvi/L*er* derived RIL population (1). Left arrows indicate that the Cvi allele increases the phenotypic value of the trait, right arrows that the Cvi allele decreases the phenotypic value of the trait. Abbreviations in brackets next to QTLs are given as reference to aid identification of trait titles used in the original publication. (**b**) QTLs mapped for inflorescence development (2) using the same Cvi/L*er* RIL population or (**c**) a RIL population derived from Columbia and L*er* (Col/L*er*) accessions. (**d**) QTLs mapped for inflorescence development in different environments (3) using Cvi/L*er* RILs or (**e**) Col/L*er* RILs. Information on the variance in a QTL affecting a particular phenotypic trait and the direction of the effect has been previously published (1–3) and is therefore not included in the figures.

Figure S3a

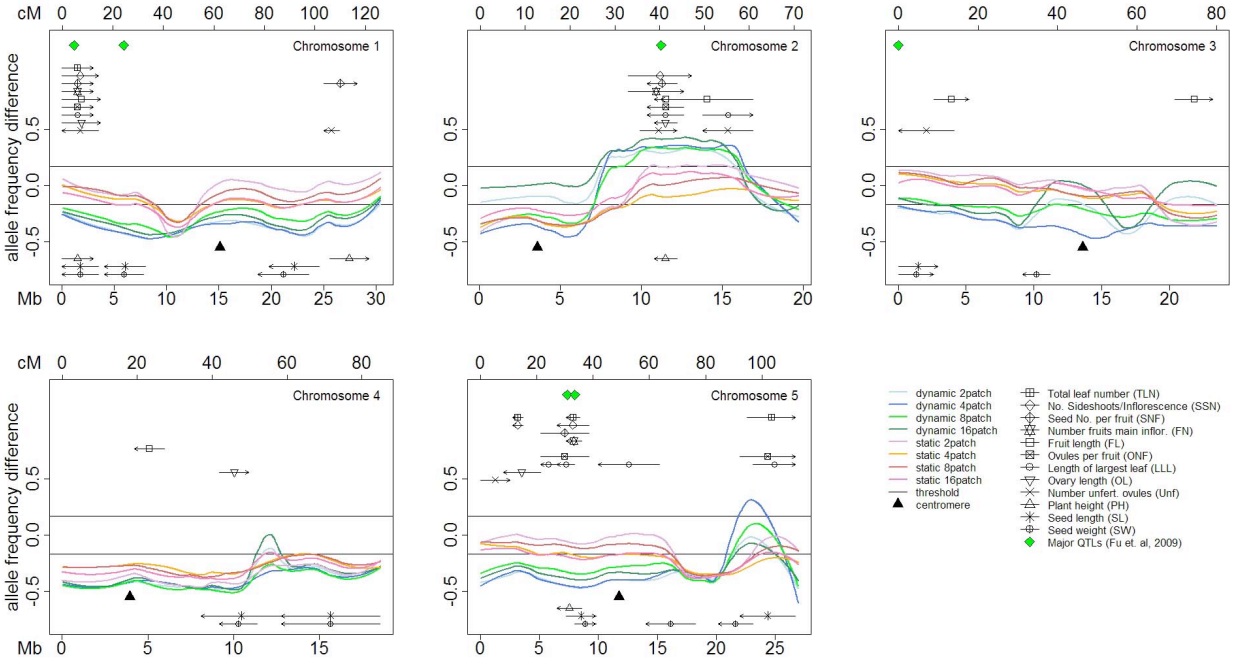

Figure S3b

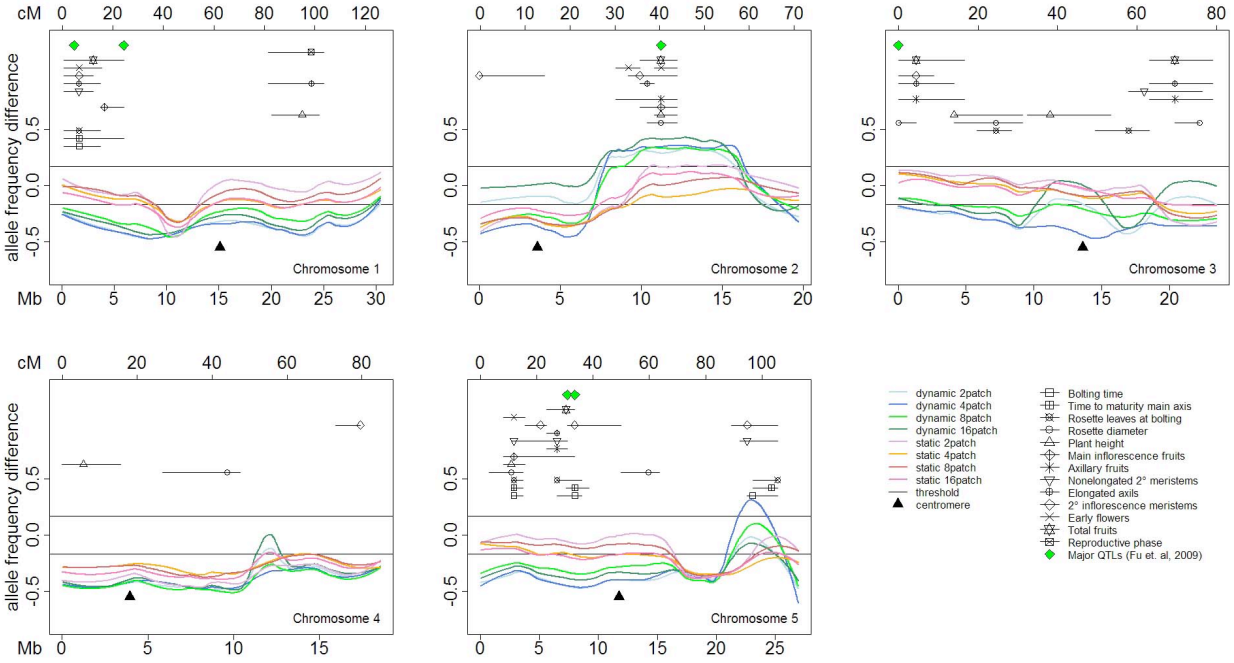

Figure S3c

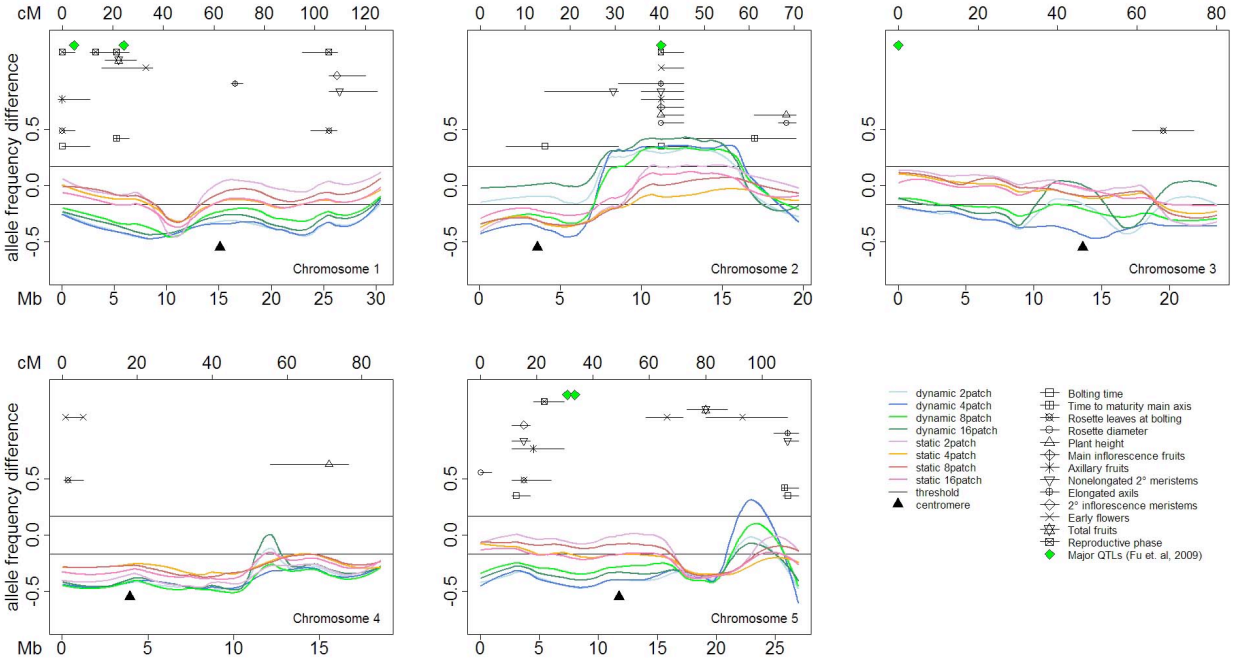
cM
Mb
allele frequency difference
Chromosome 1
Chromosome 2
Chromosome 3
Chromosome 4
Chromosome 5
dynamic 2patch
dynamic 4patch
dynamic 8patch
dynamic 16patch
static 2patch
static 4patch
static 8patch
static 16patch
threshold
centromere
Bolting time
Time to maturity main axis
Rosette leaves at bolting
Rosette diameter
Plant height
Main inflorescence fruits
Axillary fruits
Nonelongated 2° meristems
Elongated axils
2° inflorescence meristems
Early flowers
Total fruits
Reproductive phase
Major QTLs (Fu et. al, 2009)

Figure S3d

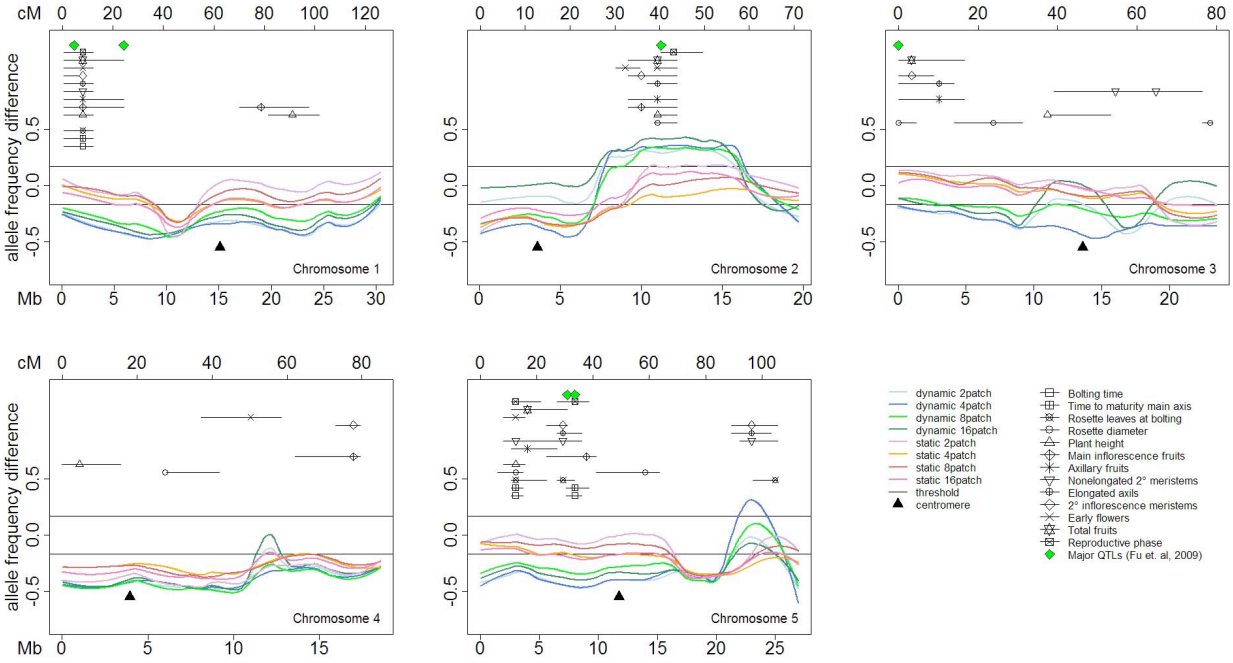

Figure S3e

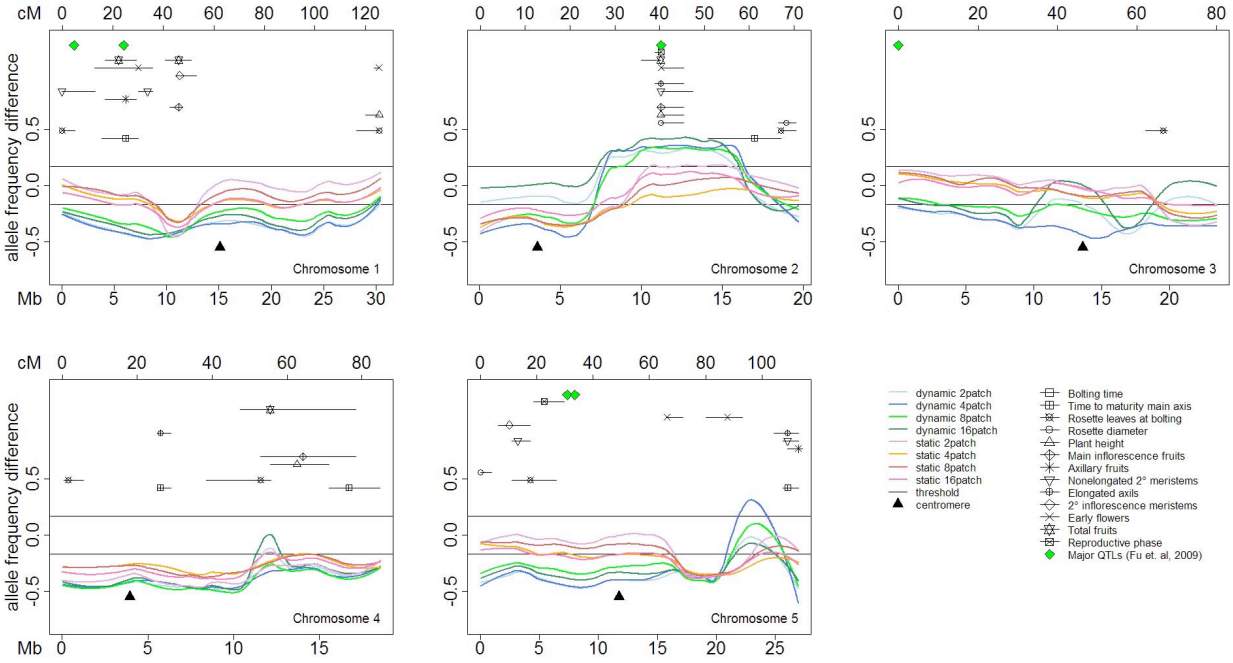

Figure S4: Expected genotypic diversity in generation 5 under drift alone. We generated a distribution for the expected genotypic diversity under drift alone for three dynamic (open triangles) and three static (open circles) landscapes. This was achieved by randomly sampling genotypes based on their frequencies in the previous generations only (in generation 1, each genotype had the same probability of being selected). The total population size was constrained to be the total number of adults observed in each landscape in each generation. The mean and 95% confidence interval are shown based on 5000 simulations per landscape. The observed genotypic diversity in each of these landscapes is also shown (solid points).

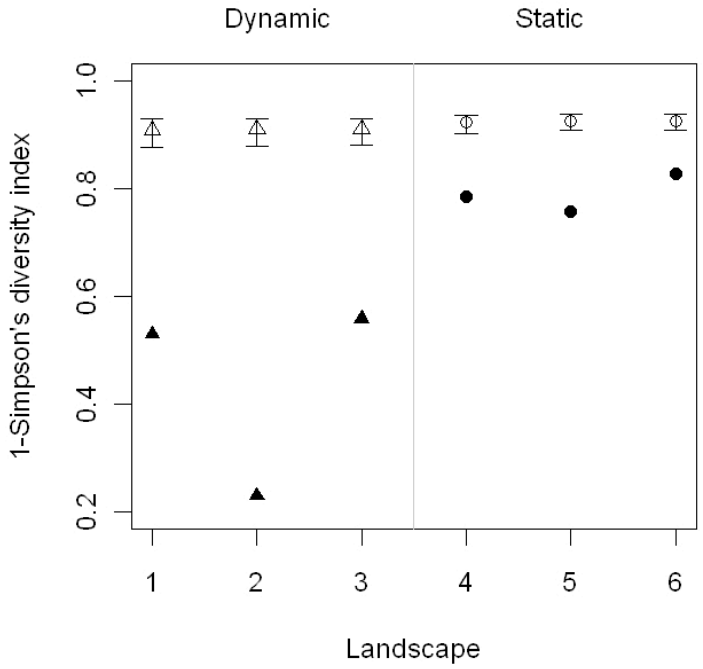

Table S1: Details of the 19 lines (4) used in the selection experiment and their abundance (based on genotyping) in static and dynamic landscapes after five generations.

Plant line	Erecta	Mass of 100 seeds (mg)	Height (mm)	% L*er* alleles	RGR (size-standardised)	No. static	No. dynamic
Cvi	0	4.45	219	0	NA	0	0
CVL60	1	5.07	133.75	32	0.339	0	0
CVL128	0	3.53	397.25	35	0.336	0	0
CVL142	1	3.03	111.75	36	0.300	0	0
CVL168	0	3.78	273.75	36	0.371	19	2
CVL37	0	4.00	277.25	52	0.356	5	1
CVL137	0	2.85	271	53	0.293	0	0
CVL158	1	4.41	193.75	53	0.389	11	0
CVL53	1	3.47	151.25	53	0.329	0	0
CVL187	1	2.50	160	54	0.333	5	0
CVL27	1	2.62	117	55	0.342	2	0
CVL135	1	3.79	168.75	59	0.320	2	0
CVL31	0	3.36	231	59	0.302	0	0
CVL19	1	2.34	183	72	0.286	0	0
CVL179	1	3.32	163.25	73	0.346	0	0
CVL39	0	2.76	403.5	76	0.334	17	55
CVL34	0	2.92	293.25	78	0.341	5	1
CVL125	0	2.54	377.75	90	0.365	13	51
Ler	1	2.44	208.25	100	0.356	39	6
Recombinants						2	2

The number of individuals belonging to the 19 ancestral lines in generation-5 populations from 16-patch static (n = 120) and 16-patch dynamic (n = 118) landscapes is shown together with the mass of 100 seeds and plant height. Lines are arranged in increasing % of L*er* alleles. Height and seed mass are based on measurements made on four individuals grown under standardised conditions. Size-standardised RGR was measured on all lines in a separate experiment (5).

Table S2. Statistical analysis: ANOVA tables of phenotypic trait analyses. Analyses were carried out using linear mixed-effect models with Landscape number (1–24) fitted as a random effect. Disturbance = static vs. dynamic.

A. The effect of experimental treatments on seedling densities ten days after sowing

	numDF	denDF	F-value	p-value
(Intercept)	1	80	678.7055	<.0001
Disturbance	1	20	8.0151	0.0103
Generation	4	80	112.7155	<.0001
log (Patch area)	1	20	0.533	0.4738
Disturbance: Generation	4	80	33.2051	<.0001
Disturbance: log (Patch area)	1	20	0.0878	0.7701
Generation: log (Patch area)	4	80	10.9737	<.0001
Disturbance: Generation: log (Patch area)	4	80	0.4308	0.786

B. The effect of experimental treatments on the fraction of seedlings surviving to adulthood through generations 1–5. The fraction Adults/Seedlings was log-transformed and analysed because in approximately 20% of cases the number of adults was actually greater than the number of seedlings, presumably because of late seedling emergence. The analysis was carried out using linear mixed-effect models with Landscape number (1–24) and patch number fitted as nested random effects. Adults were only counted in half of the experimental patches.

	numDF	denDF	F-value	p-value
(Intercept)	1	209	384.2654	<.0001
Disturbance	1	20	76.9935	<.0001
Generation	4	209	68.9743	<.0001
log (Patch area)	1	20	3.9624	0.0604
Disturbance: Generation	4	209	7.2112	<.0001
Disturbance: log(Patch area)	1	20	0.3984	0.5351
Generation: log(Patch area)	4	209	6.9988	<.0001
Disturbance: Generation: log(Patch area)	4	209	2.2829	0.0616

C. The effect of experimental treatments on the fraction of seedlings surviving to adulthood through generations 1–5 but with the number of seedlings fitted first as covariate (see also Supplementary Table 2A above). The main effect of disturbance (static vs. dynamic) is no longer significant indicating that the reduction in the fraction of seedlings surviving in static landscapes is entirely due to higher seedling densities.

	numDF	denDF	F-value	p-value
(Intercept)	1	208	447.1698	<.0001
log (Number of Seedlings)	1	208	557.7832	<.0001
Disturbance	1	20	1.429	0.2459
Generation	4	208	50.3216	<.0001
log (Patch area)	1	20	73.6682	<.0001
Disturbance: Generation	4	208	3.723	0.006
Disturbance: log(Patch area)	1	20	0.1074	0.7466
Generation: log(Patch area)	4	208	4.9454	0.0008
Disturbance: Generation: log(Patch area)	4	208	1.3303	0.2598

D. The effect of experimental treatments on the final height of generation 5 individuals grown under standardised conditions (one plant per pot)

	numDF	denDF	F-value	p-value
Intercept	1	693	3925	<.0001
Disturbance	1	20	247	<.0001
log (Patch area)	1	20	0.017	0.898
Disturbance regime : log(Patch area)	1	20	0.347	0.563

E. The effect of experimental treatments on the seed mass of generation 5 individuals grown under standardised conditions (one plant per pot)

	numDF	denDF	F-value	p-value
Intercept	1	651	1144	<.0001
Disturbance	1	20	2.98	0.0999
log (Patch area)	1	20	0.117	0.736
Disturbance regime : log (Patch area)	1	20	0.110	0.744

Table S3 A summary of the markers, the PCR primers and the resulting fragment length polymorphism for the genotyping.

Cereon/TAIR Published Name (40)	Chr	Position (bp)	PCR primer 1 (5' – 3')	PCR primer 2 (5' – 3')	Expected Ler PCR product size (bp)	Expected Cvi PCR product size (bp)
470095	1	27,359,195	CAATAGAATTTGGCTGCCGTGCCA	ATTACGTGCCTCTCTTGTCCGCTA	218	263
458557	2	4,252,182	GTCCTGGAGATGGTGGACAG	GGCAAAACCCTAATGTGGAA	389	693
448906	2	10,873,239	GATTTACATATGCCAATCCG	CTTCCGTCTCTGTCTCAAACTG	226	251
458319	2	14,388,511	TTTGAAGAGGAACCTGTGGAGCGT	CCCAGCATGGGTAAATAAGGCAGT	176	201
464890	3	7,405,294	TTGCCTCTGTGGCTGCTACTGAAT	AGTTGACCTCACACACTGAGCCAT	71	125
473983	3	11,151,654	GTTGTCAACATTCAGGTAACCAC	GTACAATGCTCATGCCTTCTCC	155	218
nga6	3	23,042,025	ATGGAGAAGCTTACACTGATC	TGGATTTCTTCCTCTCTTCAC	128	154
G3883-1.4	4	10,612,855	TGTTTCAGAGTAGCCAATTC	CATCCATCAAACAAACTCC	700	1363
457148	5	22,456,975	CACATCTGAAGCTGTGTTGCTCGT	CGCTAACGCTCTTTGGCGATCTTT	393	510

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3. Ungerer MC, Halldorsdottir SS, Purugganan MD, and Mackay TF (2003) Genotype-environment interactions at quantitative trait loci affecting inflorescence development in *Arabidopsis thaliana*. *Genetics* 165: 353-365.
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