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EFFECTS OF FOUNDING GENETIC VARIATION ON ADAPTATION TO A NOVEL RESOURCE

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Population genetic theory predicts that adaptation in novel environments is enhanced by genetic variation for fitness. However, theory also predicts that under strong selection, demographic stochasticity can drive populations to extinction before they can adapt. We exposed wheat-adapted populations of the flour beetle (*Tribolium castaneum*) to a novel suboptimal corn resource, to test the effects of founding genetic variation on population decline and subsequent extinction or adaptation. As previously reported, genetically diverse populations were less likely to go extinct. Here, we show that among surviving populations, genetically diverse groups recovered faster after the initial population decline. Within two years, surviving populations significantly increased their fitness on corn via increased fecundity, increased egg survival, faster larval development, and higher rate of egg cannibalism. However, founding genetic variation only enhanced the increase in fecundity, despite existing genetic variation—and apparent lack of trade-offs—for egg survival and larval development time. Thus, during adaptation to novel habitats the positive impact of genetic variation may be restricted to only a few traits, although change in many life-history traits may be necessary to avoid extinction. Despite severe initial maladaptation and low population size, genetic diversity can thus overcome the predicted high extinction risk in new habitats.

KEY WORDS: Directional selection, extinction, genetic diversity, niche shift, population dynamics, trade-offs.

Classical population genetic theory predicts that the amount of additive genetic variation for fitness in a population increases the response to directional selection (Fisher 1930). More recent theoretical work suggests, however, that the interaction between genetic variation and fitness is more complex. For instance, the effect of genetic variance in a fitness-relevant trait is also determined by environmental fluctuations, which can inflict a genetic load due to changing phenotypic optima (Lande and Shannon 1996). Another genetic constraint is imposed by the genetic covariance structure, which determines the effect of selection acting simultaneously on multiple traits (Blows and Hoffmann 2005). Additionally, demographic constraints can determine the importance of genetic variation during adaptation. For instance, under very strong selection—such as that experienced during the invasion of a new, extreme habitat—populations may be reduced to such low density that they are at risk of stochastic extinction before they could adapt and recover (Lande 1988). This extinction risk is especially relevant under scenarios of sudden climate change or habitat loss, when species must rapidly adapt to novel stresses to avoid extinction (e.g., Singer et al. 1993; Singer and Thomas 1996). Such strong viability selection is often termed "hard" selection, in which fitness differences are driven by survival of individual genotypes rather than competition with other genotypes for a limiting resource ("soft" selection).

In general, if the initial mean fitness of a population experiencing a sudden environmental change is low (less than one), extinction is probable unless selection on standing genetic variation increases mean fitness. However, extinction is also possible in populations with mean fitness around one, because demographic stochasticity may produce successive generations whose

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realized reproductive rates are below their mean expectation, causing population decline (Shaffer 1981; Gomulkiewicz and Holt 1995; Whitlock et al. 2003; Orr and Unckless 2008). A few recent empirical studies support these predictions, showing that initial population size greatly influences population persistence. Under strong directional selection from heat stress, *Drosophila birchii* populations derived from larger populations (thus harboring more genetic variation) persisted longer (Willi and Hoffmann 2009). Bell and Gonzalez (2009) also showed that under a novel salt stress, experimental yeast populations with greater initial size had a higher probability of evolutionary rescue, presumably because they had greater initial variation. A number of previous experiments have also shown that inbred populations with low overall genetic variation have a slower response to artificial selection (e.g., Wade et al. 1996; Reed et al. 2003). However, the predicted effects of standing genetic variation on the rate and probability of evolutionary rescue under hard selection have not been demonstrated in an experimental system.

The dynamics of founder populations under extreme environmental change (imposing hard selection) are typically density-independent, because initial fitness is very low and population growth is typically negative. The dynamics may be divided into three phases: (1) an initial decline in population size, in which populations are increasingly susceptible to deterministic or stochastic extinction (2) a transition phase, in which populations persist at near-zero growth rates but are still vulnerable to stochastic extinction, and (3) a recovery phase, when populations successfully adapt to the habitat and regain a positive growth rate and lower probability of extinction (Gomulkiewicz and Holt 1995). During these dynamics, selection can decrease population size as well as genetic variance for fitness, both of which increase the risk of extinction. Therefore, the strength of selection relative to the initial population size and genetic variation will determine the length of each phase and the mechanism and consequences of the evolutionary rescue. During the first phase, selection could favor a novel or existing beneficial allele, causing a "hard" selective sweep (distinct from hard selection discussed above) in which a single initially rare genotype is fixed in the population and genetic variation is rapidly lost. Alternatively, a previously existing deleterious or neutral allele can become selectively favored and sweep to fixation. If the initial frequency of the favored allele is sufficiently high, this can cause a "soft" selective sweep where relatively less genetic variation—particularly around the selected locus—is lost to hitchhiking (Przeworski et al. 2005). A third possibility is that a subset of previously existing genotypes can persist by chance via genetic drift. The distinction between these mechanisms is important because the subsequent transition and recovery phases depend on the amount and nature of genetic variation persisting after the initial decline. A hard selective sweep tends to rapidly erode genetic variation; genetic drift can sometimes retain some variation; and a soft selective sweep may retain much more variation depending on initial allele frequency. In sexual species, persistent variation can subsequently facilitate the rise of novel adaptive genotypes via recombination (Barton and Charlesworth 1998). Thus, if a hard selective sweep occurs early during the invasion of a novel habitat, the rate of population recovery is limited by the absolute fitness of surviving genotypes or the number of newly arising beneficial mutations (Orr and Unckless 2008). In contrast, maintenance of genetic variation during the transition phase decreases the dependency on new mutations, with the potential for a late selective sweep of ancestral and/or novel recombinant genotypes.

Which of these scenarios plays a more prominent role during adaptation after a sudden environmental change is unclear. However, they may be distinguished by measuring the rates of population decline and recovery in populations with varying degrees of founding genetic variation. In case of a selective sweep (whether hard or soft), populations carrying more copies of beneficial alleles should decline slowly and recover fast. In contrast, genetic drift will result in large variance in population performance that is uncorrelated with initial allelic composition. However, individual heterogeneity can reduce the risk of extinction from demographic stochasticity (Fox 2005). Hence populations founded with high genetic diversity should have lower variance in performance compared to genetically depauperate populations. Finally, if recombination between beneficial alleles is important for evolutionary rescue, greater allelic diversity in the population during the transition phase should promote subsequent population recovery. Here we attempt to distinguish the role of these various factors using experimental populations that were allowed to evolve in a novel habitat conferring very low initial fitness.

In a previous paper (Agashe 2009), we showed that founding genetic variation significantly decreased extinction in flour beetle (*Tribolium castaneum*) populations, both on their ancestral resource (wheat) and in habitats containing a new resource (corn, either alone or in addition to wheat). Here, we use a subset of populations from that previous study to determine the impact of genetic variation during the three phases of population dynamics in a novel habitat (see above), and specifically on subsequent evolutionary rescue. We focus on the populations reared on the stressful novel resource (corn only), which faced strong directional selection for increased fitness on the new resource. We allowed these populations to evolve on the new resource for two additional years beyond the experiments described in Agashe (2009), and determined the role of founding genetic variation as they recovered via a resource niche shift. Such shifts in resource use play a critical role in natural populations, such as mediating response to climate change (Singer et al. 1993) and ecological divergence (e.g., Singer and McBride 2010) and speciation (Schluter 2000). Hence, understanding the role of genetic variation during

Table 1. Trait variation among four experimental strains.

Strain	Fecundity $n = 36$ females		% Egg survival $n = 150$ eggs		% Larvae choosing corn patch $n = 46$ larvae
	Wheat	Corn	Wheat	Corn	
Col-2	16.25±4.8	1.5±0.65	100±0	85.7±0.03	43.48±0.07
	(5.36)	(1.97)			
Pak-3	19.8±3.08	5.4±1.29	100±0	54.45±0.04	15.21±0.05
	(4.3)	(2.78)			
Z-7	8.33±1.67	1.0±0.58	86.65±0.03	61.65±0.04	13.04±0.05
	(3.16)	(1.86)			
Tiw-5	18.6±1.54	1.4±1.17	87.35±0.03	6.75±0.02	34.78±0.07
	(3.04)	(2.65)			

Mean ± standard error for three traits are shown, with *n* individuals measured per strain per flour type (where applicable). Standard deviation for fecundity is shown in parentheses. Table modified from Agashe 2009.

resource niche shifts is important to understand and predict these phenomena.

As described in Agashe (2009), we transferred *T. castaneum* adults from their ancestral food source and habitat (wheat flour, "W") to a novel and suboptimal resource (corn flour, "C"). Experimental populations were founded with different proportions and combinations of four beetle strains that varied with respect to fitness-related traits on wheat and corn flour (Tables 1 and 2). Previous studies have documented additive genetic variation for fitness in *T. castaneum* (Hardin et al. 1967; Via and Conner 1995), and suggest that populations can successfully adapt to corn (Inouye and Lerner 1965). However, the population dynamics during this niche shift, and the genetic mechanisms responsible for it, remain unclear. To bridge this gap, we measured (1) the rate of population decline and extinction or recovery, and (2) the change in various fitness-related traits—fecundity, egg survival, larval development rate, and rate of egg cannibalism. We show that although genetic variation facilitates adaptation to novel habitats, its effect is apparent only on some of the traits that are relevant for adaptation. Additionally, our data show that even with low initial population size, sufficient founding genetic variation can allow populations to consistently escape extinction.

Methods

EXPERIMENTAL POPULATIONS

As mentioned earlier, the experimental populations used in this study were a subset of those described previously ("corn populations" in Agashe 2009). Therefore, here we only summarize key elements of the experimental design; further details can be found in Agashe (2009). We initiated experimental populations using 120 randomly picked adult beetles from stocks of four laboratory strains of *T. castaneum* (Col-2, Pak-3, Z-7 or Tiw-5). These strains differed in various fitness-related traits on wheat and corn resources (Table 1), and interbred freely after mixing in the experimental populations. We created four different levels of genetic variation (GV) using varying proportions of each of the four strains, maximizing replication at the two extreme levels of genetic variation (Table 2). Although time and labor constraints allowed us to focus on combinations of only four strains, these four were picked randomly from available stocks that were maintained under similar transfer and population sizes for ~250 generations after collection from various locations. Therefore, our populations contain a small but representative sample of available genetic variation in *Tribolium* with similar levels of within-strain variability in fecundity (standard deviations in Table 1; however,

Table 2. Experimental design.

Genetic variation (no. of strains)	No. of possible strain combinations	No. of Beetles per strain	%Beetles per strain	Replicates per combination	Total populations
Least (1)	4 (C/ P/ Z/ T)	120	100	5	20
Low (2)	6 (CP/ PZ/ ZT/ CT/ CZ/ PT)	60	50	2	12
Medium (3)	4 (CPZ/ PZT/ CPT/ CZT)	40	33	2	8
High (4)	1 (CPZT)	30	25	5	5

The four *T. castaneum* strains are denoted C (Col-2), P (Pak-3), Z (Z-7), and T (Tiw-5).

strain Col-2 has slightly greater variation for fecundity on wheat: $P = 0.043$). We do not report within-strain variability for the other traits, because eggs and larvae from different females were pooled for these assays (see below). As expected, combining the various strains led to greater initial variability (e.g., seen for fecundity on wheat, measured on females from founding populations; Fig. S1). It is important to note that this initial variability is likely to have changed during the experiment due to recombination, selection, and population size reduction.

Two replicate single-strain populations (one each of Col-2 and Z-7) were accidentally spilled during the study, decreasing the number of replicates of these strains from five to four. For the first 36 weeks of the experiment, we censused all adults, larvae, and pupae, and transferred live individuals to fresh corn flour every other week (as described in Agashe 2009). After 36 weeks, live individuals were transferred to fresh flour once every two months without census, except for a census of adults two years from the start of the experiment (described here in the Results section).

QUANTIFYING POPULATION PERFORMANCE IN THE NOVEL CORN HABITAT

We used three measures of population performance in the novel corn habitat, corresponding to the three phases of population dynamics described in the Introduction.

Rate of initial population decline

All populations declined during the first 10 weeks of the experiment (except one replicate each of strains Z-7 and Pak-3; Fig. 1), although new adults were added to most populations during this period. We estimated $t_{1/2}$, the time taken for replicate populations of each strain combination to reach half the founding population size of 120 adults. Pooling data from replicates within each strain combination, we used a generalized additive model (gam) in R (R Development Core Team 2008) to fit a nonparametric smoothing curve describing the change in population size over time (Fig. 1), using the "predict" function to estimate $t_{1/2}$. Because $t_{1/2}$ measures the net effect of birth and death events, it is an estimate of population performance during the initial decline phase in the novel corn flour. We then used an analysis of variance (ANOVA) to test for a significant effect of genetic variation on $t_{1/2}$.

Probability of population extinction

For replicate populations of each strain combination, we quantified the proportion that went extinct during the study, described in a previous paper (Agashe 2009). Alleles from each of the four strains could also affect extinction, either simply via their presence in the founding group, or as a function of their proportion in the founding group. To estimate these effects, we used generalized linear models (GLM) with binomial errors to test whether the probability of extinction depends on (1) the presence or absence of a focal strain (a binary variable); or (2) the proportion of adults from a focal strain that were used to found the initial population. Each GLM (1 and 2) was tested separately for each of the four strains.

Rate of population recovery

For populations that did not go extinct, we used nonparametric smoothing (described above) to determine the lowest population size of each strain combination, and calculated the slope of the subsequent increase in population size. This slope represents the population growth rate during the recovery phase, which is the rate of adaptation to the novel corn resource. We used Pearson's product moment correlation to test whether the slope was correlated with the degree of founding genetic variation.

FITNESS ASSAYS

(1) Fecundity assays were carried out at week two and, for surviving populations, one and two years after the start of the experiment. We randomly chose six to eight females from each population and isolated them in vials containing 1 g of either wheat or corn flour. After allowing them to oviposit for a day, we moved them to another vial containing the alternative resource, and counted the number of eggs laid in each vial. For each female, we repeated the process to get two counts of fecundity in each flour type (total 48 h per female per flour type). We calculated the proportion of total eggs laid in corn per female. Because females and the eggs they laid were exposed to wheat flour during the assay, we discarded these females rather than returning them to their source population.

(2) To calculate egg survival and larval development rate, we collected eggs obtained from the above assay (up to 10 per female) and isolated them individually (to prevent cannibalism by larvae) in 0.5-mL plastic vials containing the flour type in which they were laid. During weekly checks, we discarded dead individuals in vials, and supplied live larvae or pupae with fresh flour. We discarded eclosed adults and noted the week of eclosion to quantify the development rate from egg to mature adult. For each population, we noted the proportion of eggs that developed into mature adults to quantify % survival in corn and wheat. Due to time constraints, we measured development rate in corn and wheat and egg survival in wheat only during the two-year fecundity assay.

(3) Two years after the start of the experimental populations, we measured the cannibalism rates of beetles from the evolved corn populations and compared them with cannibalism rates in single-strain ancestral wheat populations, with five trials for each population. In each trial, we placed three males and three females in a 35-mm Petri dish with 2 g corn flour containing 5% neutral

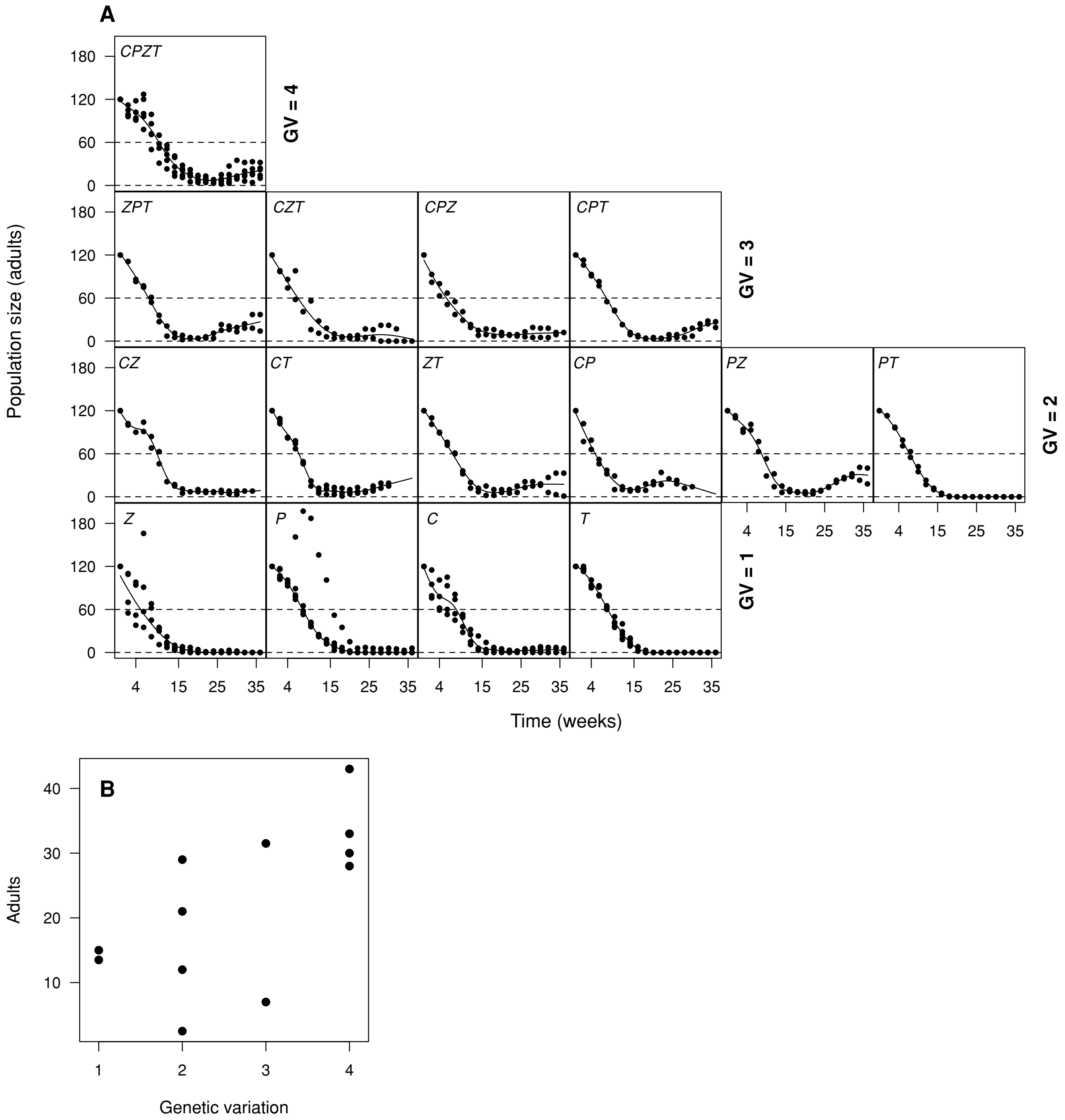

Figure 1. (A) Each panel shows data for replicate populations of a given strain combination within each level of genetic variation (GV): four combinations for single-strain (GV = 1) populations; six combinations of two strains each (GV = 2); four combinations of three-strain populations (GV = 3), and a single strain combination with the highest diversity (GV = 4) founded using all four strains. Founding strain combinations (abbreviated as in Table 2) are shown in the top left corner of each panel. Smoothed lines were predicted from a generalized additive model, excluding data from one population each from strains Z and P (bottom left panels) that increased in size during weeks 4–6. Horizontal dashed lines indicate population sizes of 0 (extinction) and 60 (corresponding to $t_{1/2}$). (B) Number of adults in persisting populations two years after the beginning of the experiment. Each point shows data for a single population of a given strain combination. For GV = 4, data for four replicate populations are shown.

red (a vital nontoxic stain that coats freshly laid eggs and stains them pink, Rich 1956). After 48 h, we sifted out stained eggs, and counted and distributed them between two 1 g layers of fresh, unstained corn flour. We returned the six adults to this fresh flour and allowed them to cannibalize for 24 h, after which we counted the remaining pink eggs (eggs laid during this period were unstained and thus could be distinguished from older, pink eggs). We calculated the per capita rate of cannibalism as the number of eggs cannibalized (No. of pink eggs remaining − No. of pink eggs introduced), divided by the number of adults.

LARVAL RESOURCE PREFERENCE ASSAY

Initially, most larvae offered a choice between wheat and corn flour patches showed an aversion to corn (Table 1). To test whether larval acceptance of the novel corn resource increased during adaptation to corn, we assayed larvae in surviving corn populations after two years (as described in Agashe 2009). Because populations now had much fewer larvae, we pooled results from larvae from multiple populations of a given strain combination to get an overall metric of larval behavioral resource preference.

Results

As expected from their low initial fitness on corn, all populations declined in size after introduction to the novel corn habitat (weeks 2–10, Fig. 1A), and 45% of populations went extinct. The egg-to-adult development time in corn flour is typically at least four weeks, and egg survival in corn flour is very low (Table 1, also see Imura 1991; Via 1991). Therefore, population size in the first few weeks of the experiment depended on the degree to which founding individuals could subsist and reproduce on corn flour. Genetic variation did not affect the time required for populations to decline to half their original size, $t_{1/2}$ ($r = 0.16$, $P = 0.57$). $t_{1/2}$ was also not correlated with the proportion of extinct populations within each level of genetic variation ($r = -0.4$, $P = 0.59$), indicating that survival and performance of the founding generation did not determine the overall probability of population extinction or persistence. Genetic variation (GV) altered within-population variability in fecundity on wheat but not corn (Fig. S1, ANOVA, effect of GV: in wheat, $P = 0.038$, in corn, $P = 0.115$). Accordingly, all populations produced similar number of larvae during the first two weeks (103 ± 2.2; effect of genetic variation: $P = 0.8$). However, as described previously (Agashe 2009), genetic variation significantly decreased the overall probability of population extinction: with increasing genetic variation (one, two, three, or four founding strains), the probability of extinction was 88.8%, 23%, 12.5%, and 0%. The presence of Pak-3, Col-2, or Z-7 founders also significantly decreased extinction (GLM: proportion extinct populations ∼ presence of focal strain; for Pak-3, $P = 0.02$; for Col-2, $P < 0.001$; for Z-7, $P = 0.03$; for Tiw-5, $P = 0.38$). However, the proportion of founding adults contributed by each strain did not affect extinction probability (GLM: proportion of extinct populations ∼ proportion of adults from focal strain; $P > 0.13$ for each strain).

Populations that did not go extinct began to recover from the initial decline (Fig. 1A), representing sufficient adaptation to the novel corn resource to permit a positive growth rate. As expected, the rate of population recovery was positively correlated with founding genetic variation ($r = 0.99$, $P = 0.01$), and translated into significantly higher adult population size of genetically diverse populations after two years (Fig. 1B; GLM, $P = 0.02$). Therefore, founding genetic variation for fitness facilitated both persistence in, and adaptation to, the novel corn habitat. During adaptation, populations with greater founding genetic variation also had higher relative fecundity in corn compared to low-diversity populations (Fig. 2A; GLM with binomial error, model: proportion eggs in corn ∼ GV * time, P [GV] = 0.77, P [time] = 0.69, P [GV × time] = 0.01). Egg survival in corn also increased significantly over time, but was unaffected by genetic variation (Fig. 2B; GLM, full model: proportion survived ∼ GV * time, AIC = −19.56; reduced model: proportion survived ∼ GV + time, AIC = −21.54, P [GV] = 0.07, P [time] < 0.001). On the other hand, egg survival in wheat did not change as a function of time or genetic variation (Fig. 2C; GLM, model: proportion

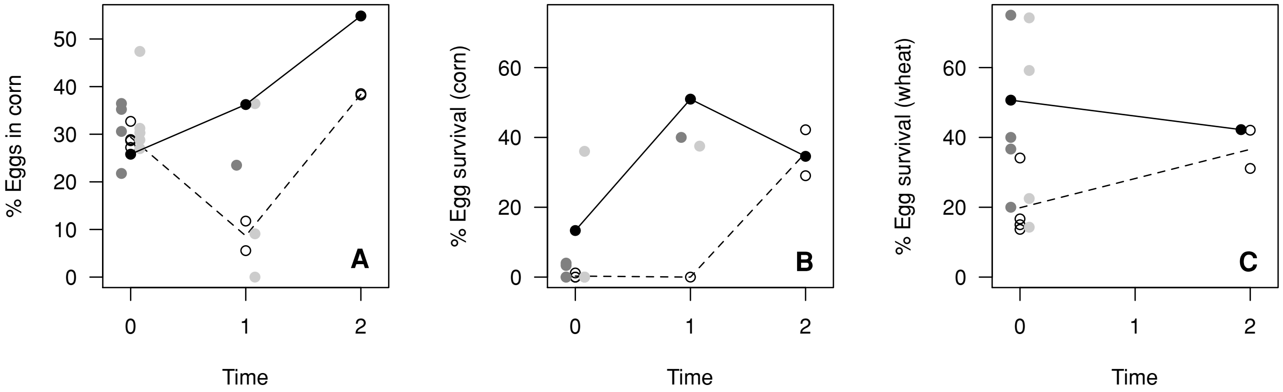

Figure 2. **(A) Relative female oviposition in corn as a function of time (years from the start of the experiment). (B and C) Egg survival to adulthood as a function of time, for eggs isolated in corn (B) and wheat (C) flour. Points, scattered slightly on the *x*-axis for clarity, are shaded to indicate increasing founding GV: empty circles show data for GV = 1 and black filled circles show data for GV = 4. Lines connect means at each time point, for GV = 1 (dashed) and GV = 4 (solid).**

Figure 3. Per capita cannibalism (in corn) of adult beetles sampled from wheat-adapted (ancestral) and corn-adapted (evolved) populations. Points are slightly scattered along the *x*-axis for clarity, and shaded to indicate increasing founding GV: empty circles show data for GV = 1 and black filled circles show data for GV = 4.

survived ~ GV × time, $P > 0.1$ for all effects). Note that average egg survival at time 0 is lower than those measured for stock populations (Table 1), likely because females from experimental populations had already been exposed to stressful corn flour for two weeks before the assay.

In *T. castaneum*, cannibalism is thought to be adaptive in nutritionally poor habitats, helping populations survive periods of large mortality (Via 1999). Therefore, we tested whether cannibalism had increased as populations evolved in the corn habitat. We found that the per capita rate of egg cannibalism in corn increased significantly from 0.1 eggs/day/adult in the ancestral wheat-adapted populations to 0.42 eggs/day/adult in the corn-adapted populations (Fig. 3; Welch two-sample *t*-test, $P = 0.003$). Comparing single-strain and four-strain corn-adapted populations (for which multiple populations were tested), we found no effect of genetic variation on the mean cannibalism rate (Welch two-sample *t*-test, $P = 0.64$).

Larval development rate in both corn and wheat increased significantly during adaptation to corn, with most eggs maturing to adulthood about a week earlier than at the start of the experiment (Fig. 4; paired *t*-test for change in mode of development time: in corn, $P = 0.03$; in wheat, $P < 0.001$). Unfortunately, due to low population size in corn we did not have sufficient larvae from each population and therefore we could not test for an effect of genetic variation on the change in development rate.

Lastly, after two years, 72% of tested larvae ($n = 18$) from corn populations chose corn over wheat flour in two-resource choice assays. This represents a significant increase in corn acceptance compared to the maximum acceptance measured before

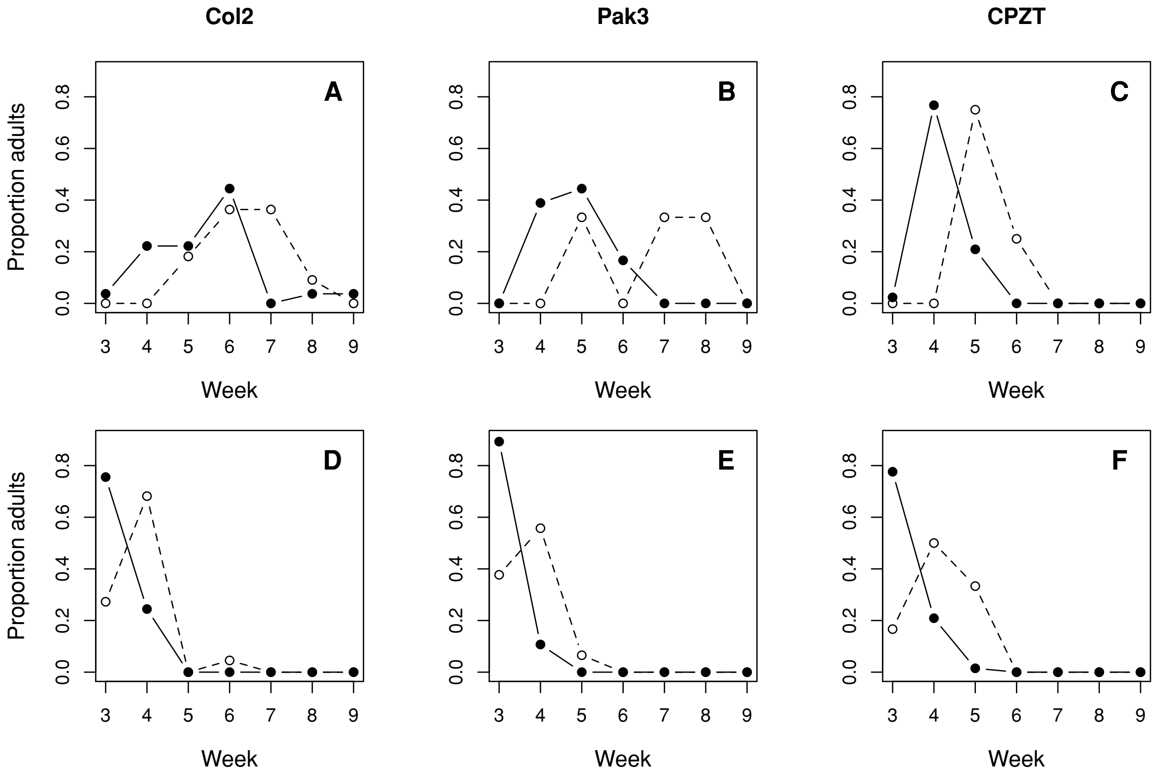

Figure 4. Initial (open circles, dashed lines) and final development rate (filled circles, solid lines) of eggs isolated from populations that evolved in corn, measured as the proportion of adults emerging each week in corn (A–C) and wheat (D–F) flour. Panels show single-strain Col-2 populations (A, D), single-strain Pak-3 populations (B, E), and high-diversity CPZT populations (C, F).

starting the experiment (43.48% for larvae of strain Col-2, $n = 46$, see Table 1; two-sample test for equality of proportions, $P = 0.038$). Therefore, during adaptation to corn, larvae in the corn populations also underwent a behavioral shift in acceptance of the novel suboptimal resource.

Discussion

In a constant novel environment, the rate of increase in fitness of an organism is proportional to the degree of additive genetic variance in fitness at that time (Fisher 1930; Price 1972). This prediction is indirectly supported by experiments showing that irradiated and hybrid *Drosophila* populations had greater population size at different temperature regimes, presumably because they carried novel mutations that increased genetic variance (Ayala 1965; Ayala 1968). Recently, Pujol and Pannell (2008) also showed that in the plant *Mercuralis annua*, reduced genetic variation at range margins was associated with reduced response to selection for pollen production. Finally, our previous work shows that under strong resource competition for an ancestral resource, founding genetic variation in *T. castaneum* populations increases the rate of dietary niche expansion onto a novel resource patch within a heterogeneous habitat (Agashe and Bolnick 2010). Here, we quantified adaptation by wheat-adapted flour beetle populations experiencing strong directional selection after sudden transfer to a novel corn habitat, that is in the complete absence of the ancestral wheat resource. Although some experimental populations went extinct, others adapted successfully to the suboptimal corn resource. In support of Fisher's theorem, we found that recovery-phase growth rate increased as a function of founding genetic variation for fitness on corn, translating into higher population size and fitness at the end of the experiment. Below we discuss the possible mechanism(s) responsible for the observed rapid adaptation to corn, and variation in rates of extinction and adaptation.

DYNAMICS OF EVOLUTIONARY RESCUE

A few mathematical models have predicted conditions in which closed populations could evolve sufficiently fast to avoid extinction in a novel environment (Gomulkiewicz and Holt 1995; Orr and Unckless 2008; Gomulkiewicz and Houle 2009). Persistence is more probable when populations start out at sizes greater than a critical extinction threshold (e.g., larger than 100). Persistence is also easier when fitness variation is largely heritable, and initial maladaptation is low. Thus, models generally predict that only weakly maladapted populations with large initial sizes can persist in novel habitats. In our current experiment, populations had a low initial size of 120 adults, which is close to the proposed extinction threshold. All populations were also strongly maladapted to the novel corn habitat. For instance, whereas the corn populations each had an average of 103 larvae within the first two weeks, identical populations maintained in the ancestral wheat resource produced ∼877 larvae in the same period (larval census data shown in Supplement 1, Agashe 2009). Furthermore, in about 7.5 weeks (less than two generations), all corn populations underwent a 50% reduction in initial population size. Despite the low starting population size and high initial maladaptation, however, most multistrain populations adapted to the novel corn habitat rapidly enough to avoid extinction. Although our experiment was not designed to directly test the models mentioned above, our results suggest that the model predictions are generally correct with respect to the importance of standing genetic variation. When populations were limited by standing genetic variation, most of them went extinct; but when starting out with large genetic variation for fitness components on corn, most adapted fast enough to avoid extinction.

The population dynamic and fitness data that we collected offer the potential to infer the roles of genetic drift, selection, and recombination during adaptation to corn (see Introduction). Adult *T. castaneum* are remarkably resistant to starvation and, in some cases, can survive a few weeks without food in isolation (Sokoloff 1977). Although the initial phase of decline in adult numbers was a net effect of birth and death, the number of new adults that emerged in most populations during this period was very small (usually less than 10), such that any differences in fecundity and egg survival were negligible relative to the overall high mortality rate. Therefore, the initial population decline reflects nonselective starvation-related mortality rather than fecundity- or egg survival-based fitness on corn specifically, which may explain the lack of a significant effect of genotype or genetic variation on the rate of population decline. Following the initial decline, however, high founding genetic variation successfully prevented extinction (Agashe 2009). Founding genetic variation also increased the rate of subsequent recovery and adaptation (Figs. 1 and 2). This positive effect of genetic variation could have resulted from selective fixation of existing beneficial alleles, and/or novel beneficial allele combinations generated by recombination among strains. Below we attempt to distinguish these mechanisms and infer the roles of selective sweeps, genetic variation, and recombination during adaptation to the novel corn resource.

Our data suggest that alleles from Col-2 and Pak-3 strains were necessary but not sufficient for consistent successful adaptation to corn. The only single-strain populations that persisted for two years were founded using adults from Col-2 or Pak-3 strains, and these strains were present in all but one surviving multistrain population (ZT). This pattern of extinction indicates that alleles from Col-2 and Pak-3 strains were necessary for persistence and adaptation in corn. Evidence that these two strains were not sufficient for corn adaptation is obtained by examining the rate of adaptation as a function of initial genotype frequencies. If fixation of alleles from either of these two strains were the sole explanation for higher survival and faster adaptation, we would expect the rate of recovery to be lower in high-diversity populations (where these strains contributed only 25% of alleles) than in medium or low-diversity populations (containing 33%, 50%, or 100% of Col-2 or Pak-3 alleles) (Orr and Unckless 2008). Instead, we observed a positive correlation between rate of recovery and genetic diversity, and found that the proportion of founding adults from each strain did not affect extinction. Thus, although the presence of Col-2 or Pak-3 genotypes reduced extinction and increased the rate of recovery, the initial frequency of these strains had no equivalent effect. On the other hand, the significant positive effect of genetic variation on population recovery rate and final population size (Fig. 1B) suggests that interactions with alleles from other strains were also important for adaptation. Therefore, it is likely that recombination between strains led to the emergence of novel genotypes that enhanced the rate of adaptation in multistrain populations.

Thus, our data strongly support an important role for recombination during adaptation in high-diversity populations. However, we have limited ability to confirm the mechanisms responsible for adaptation in low-diversity populations. For instance, it is unclear why only two specific replicate populations of Col-2 and Pak-3 managed to recover and adapt to corn, whereas all five replicates of high-diversity populations survived (Fig. 1A). One explanation is that the populations were not as genetically "identical" as intended. For instance, compared to the other strains, Col-2 had slightly greater variation for fecundity on wheat. If Col-2 and Pak-3 indeed harbored greater variability for other fitness-related traits (that we were unable to detect), sampling effects in population replicates may have caused the observed variance in extinction probability. Another possibility is that genetic or demographic stochasticity played a large role in determining the dynamics of these genetically depauperate populations, leading to successful adaptation in only a few replicates. A second limitation of our study is that without specific molecular data on the loci under selection, we cannot distinguish between hard and soft selective sweeps that may have affected adaptation in low-diversity populations. Finally, despite strong selection and sexual recombination, large linkage disequilibrium (LD) blocks from our founding strains could have persisted in our experiment, whereas natural populations typically have small LD blocks. However, many natural populations do harbor substantial LD due to migration, hybridization, and segregating inversion polymorphism; and we see a strong signal of successful recombination in our high-diversity populations. Therefore, although our results may not exactly extend to all populations (because of the way genetic variation was generated), they are broadly applicable in showing that the amount of standing genetic variation can determine the mechanism of adaptation to extreme habitats.

FOUNDING GENETIC VARIATION AND FITNESS COMPONENTS

Density-dependent egg cannibalism plays a major role in the regulation of *T. castaneum* populations (Mertz 1972; King and Dawson 1973). Although extremely high rates of cannibalism would clearly be disfavored, egg cannibalism may sometimes be adaptive due to its nutritional benefits. For instance, compared to larvae that are deprived of eggs, larvae that are allowed to eat eggs have greater fecundity as adults (Ho and Dawson 1966) and increased development time, survival, and fecundity in a suboptimal resource (Via 1999). Furthermore, cannibalism rate has been shown to have high genetic variance, with different strains cannibalizing at different rates (Stevens 1989; Via 1999). Thus, the potential exists for selection to increase the rate of cannibalism as an adaptive mechanism for survival in novel habitats. Our data show that beetles under long-term exposure to corn flour evolved higher cannibalism rates, although we did not find significant effects of genetic variation on the evolved degree of cannibalism. As population density increases further with adaptation to corn, the relative costs and benefits of cannibalism are expected to change. It would thus be interesting to test whether cannibalism rates increase or are maintained in these populations in the long term, because cannibalism cannot increase indefinitely without decreasing population growth.

A prerequisite for evolutionary trait change is the existence of heritable variation, which was present in our founding populations for multiple fitness-related traits (Table 1). However, increasing overall founding genetic variation only facilitated the increase in relative fecundity but not survival or development rate. These different effects of genetic variation may thus reflect the amount of standing additive genetic variation for each specific trait. For instance, previous work with *T. castaneum* shows that heritability for fecundity (0.5 ± 0.2) is greater than for early stage viability (0.03 ± 0.02) in wheat flour (Fernandez et al. 1995), suggesting that egg viability is largely influenced by nonadditive genetic or environmental variance. Thus, exposure to the corn habitat (i.e., environmental change) may mask genetic variation for egg viability, potentially explaining the lack of an effect of genetic variation on egg viability. Although we do not have sufficient statistical power to test for effects of genetic variation on the change in development time, previously reported estimates of heritability for development rate (0 to 0.28, Via and Conner 1995) similarly suggest that effects of founding genetic variation on the rate of change in development rate would be weak or difficult to detect.

Previous studies of adaptation in novel habitats demonstrate the existence of fitness trade-offs, when adaptation to one habitat decreases fitness in another habitat (see Hereford 2009 for a recent review). However, a previous experiment with *T. castanaeum* showed a lack of genetic correlation between development times on wheat and corn, suggesting that evolution in each environment could occur independently (Via and Conner 1995). Our experiment finds little evidence for trade-offs during adaptation: increased larval survival on corn did not alter survival on wheat. In fact, the observed adaptive increase in development rate on corn was associated with faster development on wheat. Thus, adaptation was generally beneficial across both environments.

In conclusion, we show that *Tribolium* beetles adapted to a novel resource in diverse ways, including increased per capita population growth (fitness), greater larval acceptance of the novel resource, and accelerated larval development rate. The increase in mean fitness was facilitated by founding genetic variation. Consequently, genetic variation greatly enhanced population persistence and recovery in a novel inhospitable environment, allowing populations to successfully escape extinction despite low initial population size and high degree of maladaptation. Notably, the adaptive benefits of genetic variation were apparent only for a few fitness-related traits. Other traits (development and behavioral resource choice) also exhibited adaptive changes but were uncorrelated with founding genetic diversity. Finally, our population census data suggest that the dynamics of adaptation may be driven by different mechanisms depending on founding genetic variation. These results thus increase our understanding of the role of founding genetic variation during adaptation under strong directional selection in new habitats. Although our experiments were conducted with laboratory populations, they address the fundamental dynamics of adaptation to drastically altered environmental conditions, and hence are also applicable for understanding species' responses to climate change.

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LITERATURE CITED

Agashe, D. 2009. The stabilizing effect of intraspecific genetic variation on population dynamics in novel and ancestral habitats. Am. Nat. 174:255–267.

Agashe, D., and D. I. Bolnick. 2010. Intraspecific genetic variation and competition interact to influence niche expansion. Proc. R. Soc. Lond. B 277:2915–2924.

Ayala, F. J. 1965. Evolution of fitness in experimental populations of *Drosophila serrata*. Science 150:903–905.

———. 1968. Genotype, environment, and population numbers. Science 162:1453–1459.

Barton, N. H., and B. Charlesworth. 1998. Why sex and recombination? Science 281:1986–1990.

Bell, G., and A. Gonzalez. 2009. Evolutionary rescue can prevent extinction following environmental change. Ecol. Lett. 12:942–948.

Blows, M. W., and A. A. Hoffmann. 2005. A reassessment of genetic limits to evolutionary change. Ecology 86:1371–1384.

Fernandez, A., M. A. Toro, and C. Lopezfanjul. 1995. The effect of inbreeding on the redistribition of genetic variance of fecundity and viability in *Tribolium castaneum*. Heredity 75:376–381.

Fisher, R. A. 1930. The genetical theory of natural selection. Clarendon Press, Oxford, UK.

Fox, G. A. 2005. Extinction risk of heterogeneous populations. Ecology 86:1191–1198.

Gomulkiewicz, R., and R. D. Holt. 1995. When does evolution by natural selection prevent extinction? Evolution 49:201–207.

Gomulkiewicz, R., and D. Houle. 2009. Demographic and genetic constraints on evolution. Am. Nat. 174:E218–E229.

Hardin, R. T., J. C. Rogler, and A. E. Bell. 1967. Genetic and environmental interactions in growth of *Tribolium castaneum*. Can. J. Zool. 45:139–144.

Hereford, J. 2009. A quantitative survey of local adaptation and fitness trade-offs. Am. Nat. 173:579–588.

Ho, F. K., and P. S. Dawson. 1966. Egg cannibalism by *Tribolium* larvae. Ecology 47:318–322.

Imura, O. 1991. A comparative study of the feeding habits of *Tribolium freemani* (Hinton) and *Tribolium castaneum* (Herbst) (Coleoptera, Tenebrionidae). Appl. Entomol. Zool. 26:173–182.

Inouye, N., and I. M. Lerner. 1965. Competition between *Tribolium* species (Coleoptera, Tenebrionidae) on several diets. J. Stored Prod. Res. 1:185–191.

King, C. E., and P. S. Dawson. 1973. Population biology and the *Tribolium* model. Evol. Biol. 5:133–227.

Lande, R. 1988. Genetics and demography in biological conservation. Science 241:1455–1460.

Lande, R., and S. Shannon. 1996. The role of genetic variation in adaptation and population persistence in a changing environment. Evolution 50:434–437.

Mertz, D. B. 1972. The *Tribolium* model and the mathematics of population biology. Annu. Rev. Ecol. Syst. 3:51–78.

Orr, H. A., and R. L. Unckless. 2008. Population extinction and the genetics of adaptation. Am. Nat. 172:160–169.

Price, G. R. 1972. Fisher's 'fundamental theorem' made clear. Ann. Hum. Genet. 36:129–140.

Przeworski, M., G. Coop, and J. D. Wall. 2005. The signature of positive selection on standing genetic variation. Evolution 59: 2312–2323.

Pujol, B., and J. R. Pannell. 2008. Reduced responses to selection after species range expansion. Science 321:96.

R Development Core Team. 2008. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Reed, D. H., E. H. Lowe, D. A. Briscoe, and R. Frankham. 2003. Fitness and adaptation in a novel environment: effect of inbreeding, prior environment, and lineage. Evolution 57:1822–1828.

Rich, E. R. 1956. Egg cannibalism and fecundity in *Tribolium*. Ecology 37:109–120.

Schluter, D. 2000. The ecology of adaptive radiation. Oxford Univ. Press, New York.

Shaffer, M. L. 1981. Minimum population sizes for species conservation. BioScience 31:131–134.

Singer, M. C., and C. S. McBride. 2010. Multitrait, host-associated divergence among sets of butterfly populations: implications for reproductive isolation and ecological speciation. Evolution 64: 921–933.

Singer, M. C., and C. D. Thomas. 1996. Evolutionary responses of a butterfly metapopulation to human- and climate-caused environmental variation. Am. Nat. 148:S9–S39.

Singer, M. C., C. D. Thomas, and C. Parmesan. 1993. Rapid human-induced evolution of insect host associations. Nature 366: 681–683.

Sokoloff, A. 1977. The biology of *Tribolium*. Clarendon Press, Oxford.

Stevens, L. 1989. The genetics and evolution of cannibalism in flour beetles (Genus *Tribolium*). Evolution 43:169–179.

Via, S. 1991. Variation between strains of the flour beetle *Tribolium castaneum* in relative performance on 5 flours. Entomol. Exp. Appl. 60: 173–182.

———. 1999. Cannibalism facilitates the use of a novel environment in the flour beetle, *Tribolium castaneum*. Heredity 82:267–275.

Via, S., and J. Conner. 1995. Evolution in heterogeneous environments—genetic variability within and across different grains in *Tribolium castaneum*. Heredity 74:80–90.

Wade, M. J., S. M. Shuster, and L. Stevens. 1996. Inbreeding: its effect on response to selection for pupal weight and the heritable variance in fitness in the flour beetle, *Tribolium castaneum*. Evolution 50:723–733.

Whitlock, M. C., C. K. Griswold, and A. D. Peters. 2003. Compensating for the meltdown: the critical effective size of a population with deleterious and compensatory mutations. Ann. Zool. Fenn. 40:169–183.

Willi, Y., and A. A. Hoffmann. 2009. Demographic factors and genetic variation influence population persistence under environmental change. J. Evol. Biol. 22:124–133.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:
The following supporting information is available for this article:

Figure S1. Initial within-population standard deviation in fecundity on wheat (open circles) and corn (filled circles) is shown for each level of genetic variation (GV).

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