Canalization Breakdown and Evolution in a Source-Sink System

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abstract: Understanding the process of adaptation to novel environments may help to elucidate several ecological phenomena, from the stability of species range margins to host-pathogen specificity and persistence in degraded habitats. We study evolution in one type of novel environment: a sink habitat where populations cannot persist without recurrent immigration from a source population. Previous studies on source-sink evolution have focused on how extrinsic environmental factors influence adaptation to a sink, but few studies have examined how intrinsic genetic factors influence adaptation. We use an individual-based model to explore how genetic canalization that evolves in gene regulation networks influences the adaptation of a population to a sink. We find that as canalization in the regulation network increases, the probability of adaptation to the novel habitat decreases. When adaptation to the habitat does occur, it is usually preceded by a breakdown of canalization. As evolution continues in the novel habitat, canalization reemerges, but a legacy of the breakdown may remain, even after several generations. We also find that environmental noise tends to increase the probability of adaptation to the novel habitat. Our results suggest that the details of genetic architecture can significantly influence the likelihood of niche evolution in novel environments.

Keywords: source-sink, niche evolution, genetic canalization, individual-based model, genetic assimilation.

Most species face environments that are heterogeneous in space and time, and these species may therefore frequently confront novel habitats to which they are not adapted. An important question at the interface of ecology and evolution is how species respond evolutionarily to novel environments and what factors may constrain such evolution. The ability of a species to adapt to novel unfavorable environments may determine whether the species' geographical range is static or expanding (Kirkpatrick and Barton 1997; Case et al. 2005), whether the species can persist in degraded habitats (With 2002), and whether pathogenic species can invade new hosts (Antia et al. 2003), among other ecological phenomena. The specific type of novel environment that we examine in this article is a sink habitat (Holt 1985; Pulliam 1988). A sink is defined as a low-quality habitat where the discrete-time growth rate of a population in the absence of dispersal is less than 1, and the population becomes deterministically extinct without recurrent immigration from a source population. A source habitat, by contrast, is a high-quality habitat where the growth rate of a population when rare is greater than 1. There are many examples of natural populations with source-sink structure (e.g., Breininger and Carter 2003; Breininger and Oddy 2004; Johnson 2004; Caudill 2005).

One can define a species' niche as the combination of biotic and abiotic factors that allow the species' growth rate to be greater than 1. Therefore, a source is a habitat with conditions within the species' niche, and a sink has conditions outside the niche (Holt and Gaines 1992). Source-sink dynamics thus emerge from the demographic interplay of populations inside and outside of the species' niche; analysis of adaptive evolution in such landscapes may consequently shed light on niche evolution. Previous theoretical studies have illuminated how evolution in a sink can lead ultimately to adaptation and persistence in the sink (Kawecki 1995, 2000*a*; Holt 1996; Holt and Gomulkiewicz 1997; Gomulkiewicz et al. 1999; Holt et al. 2003). When such evolution occurs, the niche has evolved. Niche evolution has obvious implications for species persistence in degraded or fragmented landscapes (With 2002). Similarly, the realized specificity of pathogens to their hosts may be determined by constraints on evolution of populations in sink habitats, where "habitat" in this sense may be a low-quality host species (Antia et al. 2003).

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On a broader scale, if the area within a species' geographical range is considered a source and the area outside of that range a sink, evolution in sinks at the border may allow the expansion of the species' range (Griffith and Watson 2006). This process may be of particular importance in determining which introduced species are able to expand their geographical ranges and become invasive (Lee 2002; Holt et al. 2005*a*).

Most studies of source-sink evolutionary dynamics have focused on the influence of extrinsic ecological factors in the process of niche evolution, such as temporal variation in sink harshness (Holt et al. 2004*b*), density dependence (Gomulkiewicz et al. 1999), interspecific interactions (Case et al. 2005), or asymmetries in dispersal rates (Kawecki 2000*a*; Kawecki and Holt 2002). Moreover, most work to date has assumed that the genetic basis of the trait undergoing selection either is a single locus or involves additive variation at multiple loci, each of which has a small effect on a quantitative trait (e.g., Kawecki 1995, 2000*a*; Ronce and Kirkpatrick 2001; Holt et al. 2004*b*). Little attention has been given to genetic architectures other than that of standard quantitative genetics. In this article, we will examine a quite different kind of genetic architecture, one that includes the intrinsic factor of genetic canalization of development arising from a gene regulation network.

Genetic canalization is the suppression of phenotypic variation caused by mutations (Waddington 1942). There has been a great deal of interest recently in how canalization evolves (e.g., Wagner 1996; Wagner et al. 1997; Eshel and Matessi 1998; Rice 1998; Kawecki 2000*b*; Flatt 2005; Proulx and Phillips 2005). Previous theoretical studies have suggested that genetic canalization can arise in individuals with epistatic gene interactions simply through selection for developmental stability (Siegal and Bergman 2002; Bergman and Siegal 2003). Genetic canalization may influence source-sink evolutionary dynamics by impacting evolvability (Houle 1992; Flatt 2005), breaking down to release hidden genetic variation (becoming "decanalized"; Layzer 1980; Rice 1998), or interacting with extrinisic factors such as environmental noise to determine the amount of variation exposed to selection. Because the genes within a gene network interact in highly nonlinear ways to determine an individual's phenotype, there may be different selective forces at work during adaptation in a sink environment than are recognized in more classical quantitative genetic approaches. By exploring the influence of genetic canalization on source-sink dynamics, we may gain a clearer understanding of how developmental constraints influence the process of niche evolution.

Methods

To model evolution in a source-sink system, we created an individual-based model similar to the model of Holt et al. (2003). We assume that a population of individuals persists in a source habitat where they are well adapted and at evolutionary equilibrium. In each generation, individuals from the source migrate to the sink, where they are maladapted. Individuals are maladapted to the sink in our model because the phenotypic optimum in the sink is sharply different from that in the source, and therefore individuals adapted to the source have a considerably lower fitness in the sink. We do not allow back-migration to the source; thus, we are modeling a "black-hole" sink (sensu Holt and Gaines 1992). Individuals are haploid and reproduce sexually, with B offspring per adult ($B = 4$ in all model runs shown here; see the appendix, available in the online edition of the *American Naturalist*, for a discussion of effects of varying fecundity); for each offspring, its parents are chosen randomly from all adults. Ten individuals are initially introduced to the source, and for the first 400 generations of the simulation, individuals occur only in the source; this initial phase permits the source population to become canalized to the source habitat. Thereafter, I individuals migrate from the source to the sink each generation after reproduction has occurred but before density-dependent selection occurs in either habitat. If there are more than K individuals in a habitat after selection, only K individuals are randomly chosen to reproduce the next generation. This creates density dependence in both the source and the sink, with a "ceiling" carrying capacity. We assume there is no density dependence when the population is below this ceiling. Because the sink population is initially maladapted, its numbers are typically well below K. Simulations were run for 10,000 generations.

Our model differs from that of Holt et al. (2003) in our implementation of the genotype and phenotype of each individual in the model. Holt et al. (2003) followed the protocol of Burger and Lynch (1995) in assuming that trait loci combine additively to determine the value of a single quantitative trait (e.g., body size). Here, we instead base the genotype and phenotype of individuals on the model of genetic canalization developed by Siegal and Bergman (2002) and Bergman and Siegal (2003), who were in turn inspired by the model of Wagner (1996).

In our model, a network of transcriptional regulation genes describes the genotype of an individual, and each gene has the potential to regulate every other gene in the network. The phenotype of an individual is a vector of expression levels of each gene in the network; this vector changes during development and must reach equilibrium within a given time frame for individuals to successfully develop into reproductive adults. The equilibrium phenotype then determines adult survival in a given environment. This model can be thought of biologically as a network of genes, each with upstream enhancer regions that

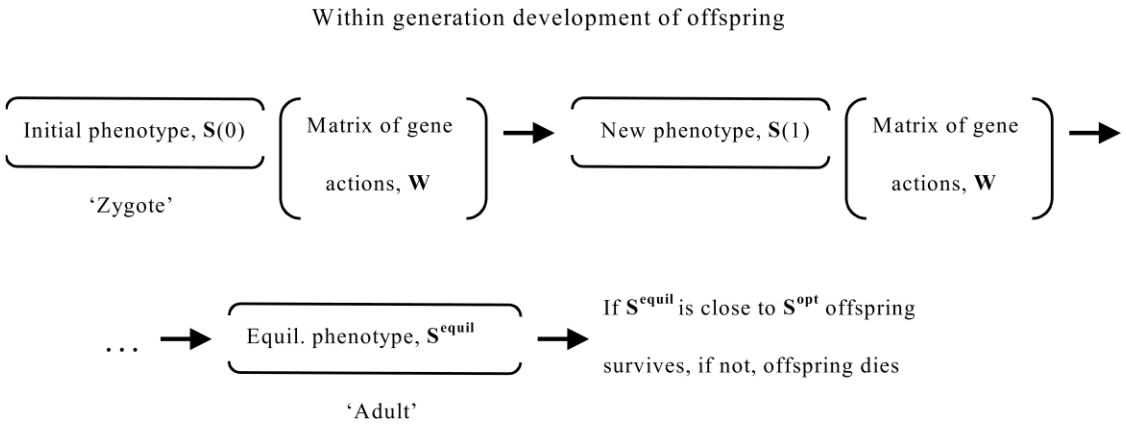


Figure 1: Diagram of within-generation development of an offspring from zygote to adult. The phenotype vector of the individual at time t, $\mathbf{S}(t)$, interacts with the matrix of gene actions, $\mathbf{W}$, to produce the new phenotype vector at time $t + 1$, $\mathbf{S}(t + 1)$. This proceeds until the phenotype vector goes to equilibrium or until 100 time steps have been reached; if equilibrium is not reached, the individual dies. Selection acts on the equilibrium phenotype to determine whether the offspring is part of the next generation. This second component of selection acts differently in the source and sink habitats.

are potentially regulated by the gene's own gene product as well as the gene products of the other genes in the network. A gene's enhancer regions and the amount of corresponding gene products produced by the genes in the network determine both that gene's strength and direction of regulation.

Simple gene network models have been successful in predicting the trajectory of gene expression levels in several biological organisms (Albert and Othmer 2003; Li et al. 2004; Ma'ayan et al. 2005). Thus, the "wiring" of a gene network may be the most important factor in determining the expression levels of genes regulated by that network (Bornholdt 2005). Siegal and Bergman (2002) and Bergman and Siegal (2003) used their model of gene regulation to explore how the complexity of the regulation network influenced genetic canalization. Our interest instead is in how genetic canalization influences niche evolution. We thus placed the Siegal-Bergman framework for individual development into the demographic context of the source-sink system examined by Holt et al. (2003) and others (e.g., Kawecki 1995). We therefore explore how the complexity of genetic wiring may influence niche evolution. We compare our model to other models of canalization in "Discussion."

We simulate the development of an individual by starting with an initial concentration of each gene product that is far from equilibrium; we then use a set of coupled difference equations to allow the phenotype of an individual to develop to equilibrium (following Siegal and Bergman 2002). Each individual in our model has n genes (we used $n = 10$ in all runs reported here), and each of those genes has the potential to regulate every other gene. We define the genotype of an individual by its $n \times n$ gene regulation matrix ($\mathbf{W}$). Each row i of the $\mathbf{W}$ matrix describes a different gene. Each column therefore describes the effects of a given gene on itself and all of the other genes in the matrix. The complexity of the regulation matrix (denoted by c) is the percentage of nonzero w_{ij} elements in the $\mathbf{W}$ matrix. We initially determine the value of each nonzero w_{ij} element by drawing from a normal distribution with mean of 0 and variance of 1. The nonzero w_{ij} elements are randomly placed in the matrix and could be either positive or negative. Any nonzero w_{ij} element on the diagonal represents autoregulation of the gene by its gene product (e.g., Sucov et al. 1990; Simpson et al. 2003; Verma et al. 2006). All of the other w_{ij} elements are set to 0, meaning they have no effect on gene i. A w_{ij} element that is initially set to 0 remains at 0 throughout the simulation.

The time-dependent phenotype of an individual, $\mathbf{s}(t)$, is a vector of the gene expression level of each gene at time t, where each expression level is denoted by $s_i(t)(i = 1, \ldots, n)$. The trajectory of the phenotype vector describes the process of development within an individual. Figure 1 shows a diagram of offspring development within a single generation. This developmental trajectory is described by iterating a set of coupled difference equations:

$$s_i(t + 1) = f\left[\sum_{j=1}^{n} w_{ij} s_j(t)\right], \tag{1}$$

where $f(x) = 2/(1 + e^{-ax}) - 1$ is a sigmoidal function with steepness determined by parameter a. We use a very steep sigmoidal function ($a = 100$) to ensure that the gene expression level for each gene is, to a very close approximation, equal to $+1$ or -1 (i.e., each gene is either completely activated $[+1]$ or repressed $[-1]$). We iterate this process until $\mathbf{s}(t)$ reaches an equilibrium, $\mathbf{s}^{\text{equil}}$, where $\mathbf{s}(t) = \mathbf{s}(t + 1)$ (to within error defined by machine accuracy) within a given time frame. The first iteration uses an initial phenotype vector, $\mathbf{s}(0)$, that is the same for both the source and sink populations and can be considered to be the initial phenotype of the individual as a zygote. The number of iterations required until $\mathbf{s}(t)$ reaches equilibrium is defined as the path length of development for that individual. If the path length is greater than 100 (which usually indicates that $\mathbf{s}(t)$ will not reach an equilibrium within a reasonable time horizon), then we consider the individual to be developmentally unstable and remove it from the simulation (i.e., it dies). Thus, selection acts on development to achieve a stable phenotype by a certain point in the life cycle.

In addition to selection on developmental stability, we assume that the adult phenotype influences survival in a habitat-specific way. The optimal phenotype for the source habitat ($\mathbf{s}^{\text{opt}}$) is set equal to the $\mathbf{s}^{\text{equil}}$ produced by the first stable individual generated at random in the source. Given that the offspring has developed successfully, its survival probability in the source is defined as

$$F\left(\mathbf{s}^{\text{equil}}\right) = e^{-D(\mathbf{s}^{\text{equil}}, \mathbf{s}^{\text{opt}})/\sigma}. \quad (2)$$

Here, $D(\mathbf{s}^{\text{equil}}, \mathbf{s}^{\text{opt}}) = \sum_{i=1}^{n} (s_i^{equil} - s_i^{opt})^2/4n$ gives the phenotypic distance between the equilibrium phenotype of an individual and the optimal phenotype in a habitat (approximately the fraction of gene products at which they differ); the parameter σ determines the strength of selection. We used a relatively strong strength of selection ($\sigma = 0.1$) in all of the simulation runs that we performed. We include the offspring in the next generation if the fitness component defined by equation (2) is greater than a random number taken from a uniform distribution between 0 and 1. Our model thus incorporates two aspects of selection acting sequentially in a life history: selection against developmental instability and stabilizing selection around $\mathbf{s}^{\text{opt}}$.

Reproduction occurs by randomly choosing two haploid individuals in the source and then producing a haploid offspring by randomly choosing rows in $\mathbf{W}$ from each parent with equal probability. This procedure describes haploid parents producing a diploid zygote, with meiosis occurring before development of the offspring, and it allows independent assortment of parental genes into the offspring. Choosing parents for reproduction occurs with replacement (which prevents an Allee effect when population numbers are small). After meiosis, each nonzero w_{ij} element has a chance of mutating with probability $0.1/cn^2$. By making the mutation rate dependent on c, we are assuring that the per-genome mutation rate is the same for all individuals, independent of their genetic complexity. If mutation occurs, the w_{ij} element is replaced with a number drawn from a normal distribution with mean of 0 and variance of 1. Although the probability is very small, a w_{ij} element could mutate to a value of 0. If this occurs, the w_{ij} value remains 0 and a new value of c is calculated for that particular matrix (in our simulations, this very rarely occurs). After mutation, development of the zygote begins.

Reproduction, development, and selection are implemented using the same protocol in the sink. However, we assume that the optimal phenotype in the sink differs from that in the source. We create the optimal sink phenotype by setting it equal to the optimal source phenotype, but we then reverse the sign of h randomly chosen gene products. In other words, h genes that are activated in the optimal source phenotype are repressed in the optimal sink phenotype or vice versa. In this way, as h increases, the sink becomes harsher for individuals from the source (as measured by survival; eq. [2]).

This means of defining the initial degree of maladaptation in the sink differs sharply from the quantitative genetic approach used by Holt et al. (2003, 2005*b*), who assumed additive affects of different loci on a single trait. In our model, the gene products that have different phenotypic optima in the sink may be influenced by every other gene, implying strong epistasis. Our model further differs from the models of Holt et al. (2003, 2005*b*) in that evolution in our model does not occur directly on an observable ecological phenotype but instead occurs on the equilibrium output of a gene regulation network. However, evolution of developmental gene regulation networks appears to have been important for adaptation and speciation in several species and may even be important in the evolution of the animal body plan (reviewed in Davidson and Erwin 2006). Thus, the outputs of gene regulation networks may be frequent targets for evolution in novel environments.

To determine the average canalization of individuals within a population, we estimate the average sensitivity to mutation of each individual within the population. Sensitivity to mutation is inversely related to genetic canalization (Wagner et al. 1997). We estimate sensitivity by randomly mutating one of the nonzero w_{ij} elements within an individual's genotype and then measuring the phenotypic distance between the unperturbed and perturbed phenotype vectors at developmental equilibrium (following Siegal and Bergman 2002). An individual's sensitivity is averaged over 10 perturbations to its genotype, and this

sensitivity is then averaged across all individuals in the population to arrive at a measure of average canalization.

Previous research (Siegal and Bergman 2002; Bergman and Siegal 2003) has shown that within a single population of reproducing individuals, both path length and sensitivity to mutation decrease with evolution, even though there is no direct selection on path length or sensitivity to mutation. Sensitivity to mutation tends to decrease because selection for developmental stability also produces robustness to mutations. Path length tends to decrease because mutations are more likely to have deleterious effects in individuals with long paths (Wagner 1996). Furthermore, the decrease in sensitivity to mutation and path length is greater with increasingly complex gene regulation networks. Thus, it was found that canalization tends to increase over time, and the larger the c value defining complexity in the regulatory matrix, the greater the canalization becomes. By letting individuals in the source evolve for 400 generations in our model before beginning immigration to the sink, we allow the source population to become genetically canalized to the source habitat. (Genetic canalization often continues to increase slightly in the source even after 400 generations, but these later changes in canalization are usually relatively small.)

We explored the model by first determining the cumulative probability of adaptation to the sink habitat per generation, given different levels of harshness in and immigration to the sink habitat. We also examined the dependence of adaptation rates in the sink on our measure of network complexity, c. We did this by running the model 100 times for each set of parameter values and recording the number of generations after immigration began until the sink population first reached 50 individuals. This number was chosen because in practice, after the sink population reaches 50 individuals, the population is sufficiently adapted to the sink habitat that it can persist without further immigration from the source. If the sink population did not reach 50 individuals in 10,000 generations, the population was considered to have never adapted to the sink environment.

To determine how canalization influences the probability of adaptation to the sink, we assessed the average path length and sensitivity to mutation for individuals in both the source and sink during adaptation to the sink habitat. This procedure was performed for four values of c, and the model was run 100 times for each value of c. We then compared the average path length and sensitivity to mutation in the same populations after 10,000 generations.

Variation in the environment at the time of zygote formation could lead to chance differences among individuals with the same genotype. To examine the impact of this component of environmental noise, we randomly perturbed the initial gene expression vector, $\mathbf{s}(0)$, of individuals in both the source and the sink. We simulated low noise by perturbing an expression level in $\mathbf{s}(0)$ of an individual with probability 0.01; high noise was simulated by perturbing an expression level in $\mathbf{s}(0)$ with probability 0.1. If an expression level was perturbed, a number taken from the zero-mean normal distribution, with standard deviation 0.01, was added to the expression level. Individuals in both habitats had a fixed probability of being perturbed each generation. We compared the probability of adaptation in the sink with environmental noise with the probability without noise.

Results

Time series of the source population show that numbers in the source quickly increase (the increase is so fast that it is difficult to see in fig. 2*A*, 2*B*). In the sink, however, numbers typically stay small for many generations after immigration from the source begins, but then they quickly increase to a plateau. Sometimes, several such plateaus emerge before the sink population is sufficiently adapted to reach the same population size as the source (e.g., fig. 2*A*; in this time series and that of fig. 2*B*, the population is censused after reproduction and selection but before density dependence acts, so the presence or absence of plateaus during the course of adaptation to the sink can be more readily seen). Once adaptation has occurred, the sink population can persist even if immigration is stopped; thus, there has been evolution in the species' niche.

This temporal pattern in abundance can be understood by examining figure 2*C*, which shows the mean phenotypic distance between individuals in the sink and the sink optimum, for the same population as in figure 2*A*. In this example $h = 2$; thus, there are differences between the optimal phenotypes in the source and the sink at two loci, and the initial mean phenotypic distance of individuals to the sink optimum is 0.2 (i.e., h/n). After an evolutionary lag, the population in the sink adapts to one of the gene expression levels that differs from the source level, and the mean phenotypic distance to the sink optimum decreases to about 0.1. This initial adaptation allows the population in the sink to increase to a new plateau. When the population in the sink then adapts to the second, differing gene expression level, the mean phenotypic distance to the sink optimum decreases to 0, and the sink population numbers increase to the same numbers as the source.

The pattern of adapting to the sink environment in several discrete, punctuated steps was not always observed. Figure 2*D* (corresponding to fig. 2*B*) shows that it was possible for a population in the sink to adapt to more than one differing optimal gene expression level almost simultaneously. Following a long period of sustained low

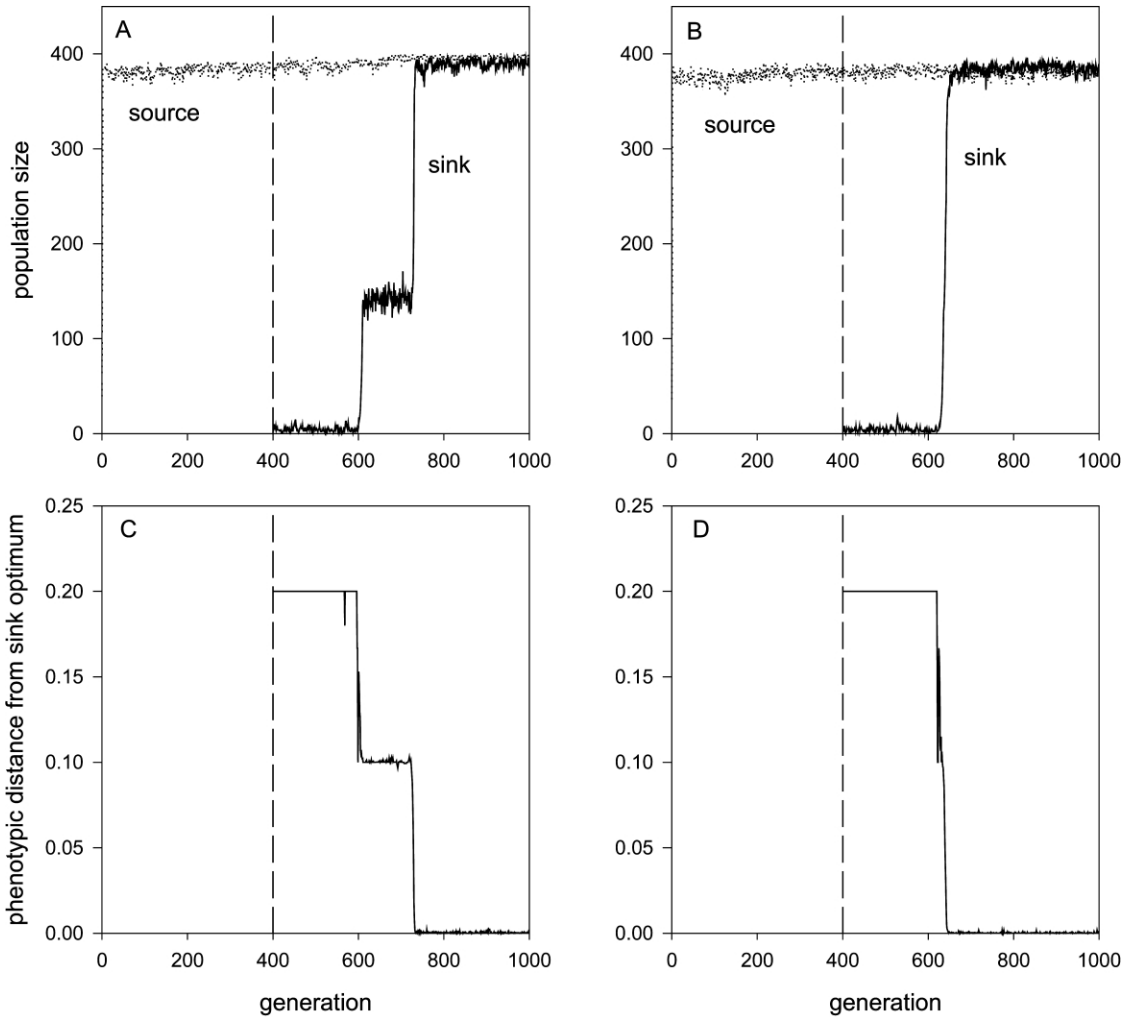
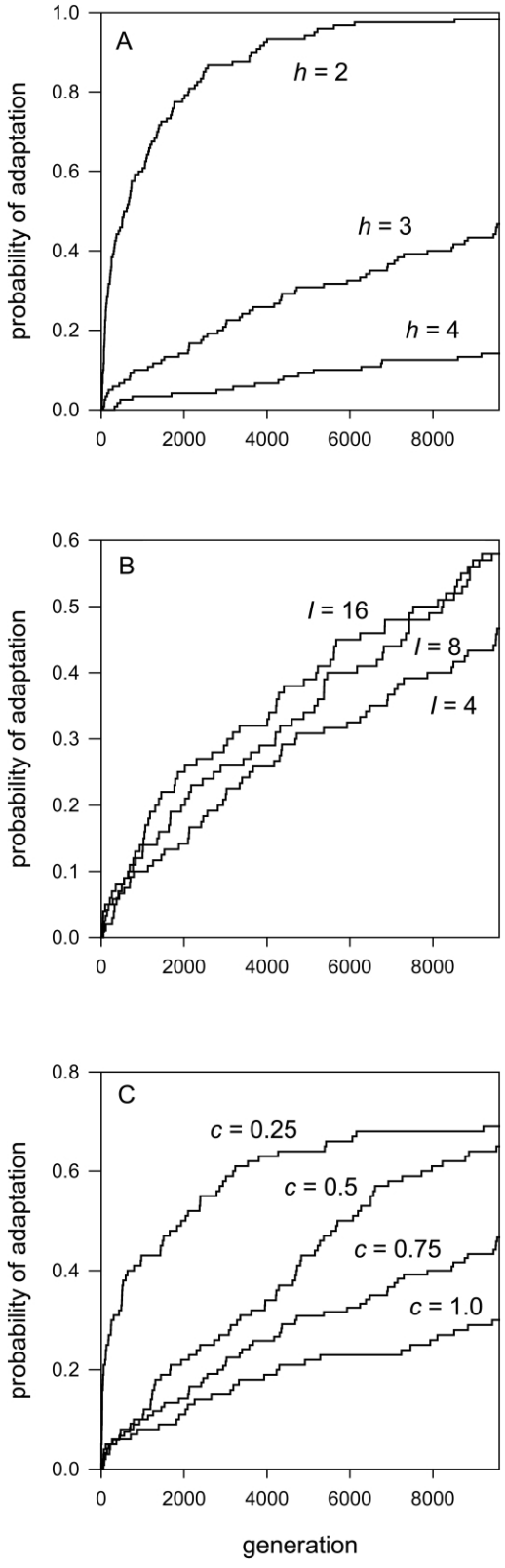


Figure 2: *A*, *B*, Representative time series of population size in the source (*dotted lines*) and sink (*solid lines*). Population size in the sink can increase in several steps or one punctuated jump. Population sizes are shown after reproduction and selection but before density dependence acts. *C*, *D*, Mean phenotypic distance of individuals in the sink from the sink optima in *A* and *B*, respectively. In each panel, the dashed line indicates when migration began to the sink. The parameters are $h = 2$, $c = 0.75$, $K = 100$, and $I = 4$.

abundance and maladaptation, the population in the sink reached the same size as the source in essentially a single step (fig. 4*A* shows another example). The ability of the sink population to increase to the same size as the source population in a single step decreases with increasing sink harshness. We never observed simultaneous adaptation to four or more differing optimal expression levels; thus, several population plateaus were usually observed when adaptation to the sink occurred and harshness was relatively high.

As in previous studies (e.g., Ronce and Kirkpatrick 2001; Holt et al. 2005*b*), we found that the probability of adaptation to the sink decreased sharply with increasing harshness of the sink habitat (fig. 3*A*). In the example shown, the fitness of an individual with the source optimal phenotype in a sink with $h = 2$ is 0.135; in a sink with $h = 4$, the fitness is 0.018. In the latter case, even after 10,000 generations, more than 80% of the populations had not adapted to the sink. There is no absolute genetic constraint on adaptation in this model but instead a kind of quasi-equilibrial stasis. Interestingly, when h is very high (e.g., $h = 9$), the probability of adaption to the sink increases relative to a slightly less harsh sink (e.g., $h = 7$). This occurs because it is possible for mutations to arise that cause all of an individual's gene expression levels to flip, with activated genes becoming repressed and repressed genes becoming activated. If this occurs in a sink where $h = 9$, then the mutant individual differs from the



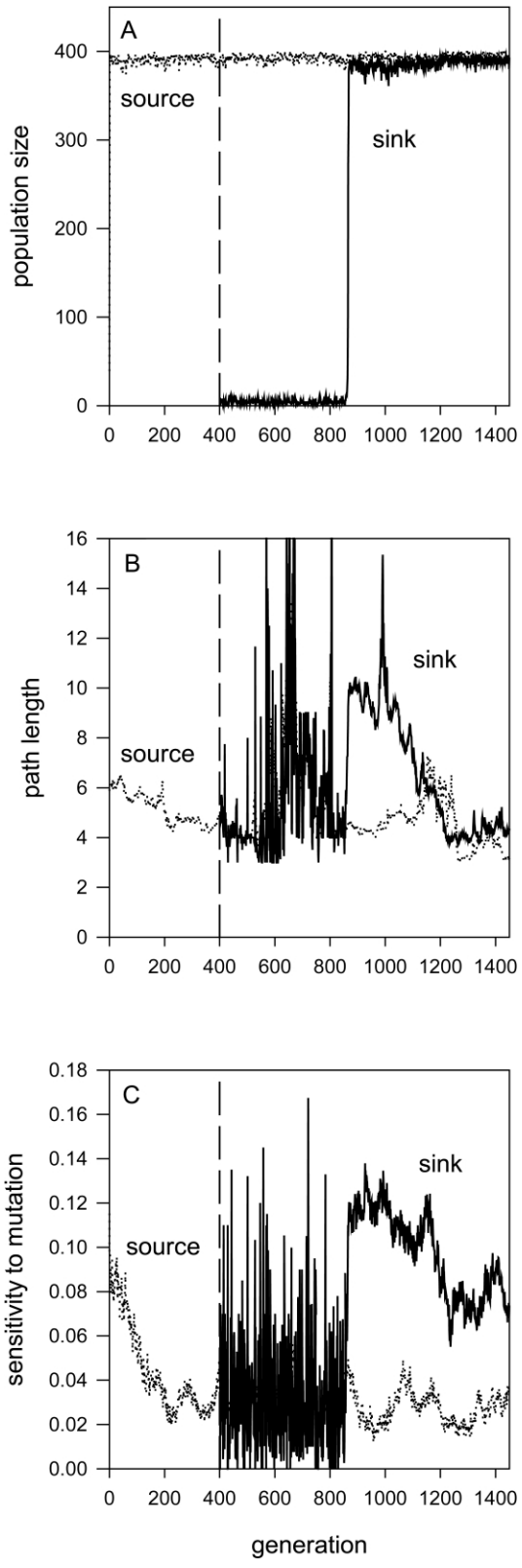
optimal sink phenotype at only one locus, and the probability of adaptation to the sink thus increases.

The probability of adaptation increased with increasing immigration from the source (fig. 3*B*). The effect of immigration on adaptation is stronger than observed in multilocus additive genetic models without density dependence (Holt et al. 2005*b*). As Gomulkiewicz et al. (1999) explained for a one-locus model, this pattern can be expected if immigration provides a conduit for genetic variation to be supplied to the sink. Similar to a result reported by Holt et al. (2003), the probabilities of not adapting to the sink as a function of time when plotted on a semilog plot are approximately linear (fig. A1), suggesting that the probability of adapting to the sink habitat is roughly constant per unit time. Several of these results hint that adaptation to the sink may be the result of mutations of relatively large effect. To assess this interpretation in detail, we would need to follow the phylogeny of each new mutation produced within the source and sink populations; however, the large population sizes and number of generations used in our simulations made this computationally prohibitive for this initial study, so we defer analysis of this issue to future reports.

A new conclusion from our model is that the complexity of the gene regulation matrix had a large effect on the probability of adaptation to the sink (fig. 3*C*). Given a relatively harsh sink ($h = 3$), populations with less complex regulation networks had a much higher probability of adapting to the sink over the defined time period than did populations with more complex regulation networks. This occurred, we suggest, because complex regulation networks had much greater canalization to the source optimum phenotype than did less complex regulation networks, and so they expressed less phenotypic variation upon which selection could act. Greater canalization and reduced phenotypic variability in the source population act to reduce the probability of adaptation to the sink habitat.

The scope for adaptation via selection on survival is constrained by mutation rate and fecundity. As we show in figure A2*A*, A2*B*, the probability of adaptation decreases with decreasing mutation rate and increases strongly with

Figure 3: *A*, Probability of a population becoming adapted to the sink habitat as a function of time (since start of immigration) for different levels of sink harshness *h*. Note that adaptation involves changes that permit persistence in the sink environment, that is, niche evolution. *B*, Probability of adaptation for different immigration rates *I*. *C*, Probability of adaptation for different complexities *c* of gene regulation network. The model was run 100 times for each different parameter in each panel. Parameters are $c = 0.75$ in *A* and *B*, $I = 4$ in *A* and *C*, and $h = 3$ in *B* and *C*. $K = 100$ in all panels.



B. The qualitative pattern of adaptation, however, is the same as shown in figure 3*C*, with a lower probability of adaptation at higher *c*. We also explored whether a mutation rate that is independent of *c* alters the pattern of adaptation in the sink. To assess this, we set the w_{ij} mutation rate equal to $0.1/n^2$, thereby giving individuals with greater genetic complexity a larger per-genome mutation rate. We found that as gene regulation complexity increased, the probability of adaptation in the sink still decreased (fig. A2*C*). Thus, even with a much higher per-genome mutation rate, the canalization that emerges from greater genetic complexity continued to reduce the probability of adaptation in the sink.

We can explore the impact of canalization by examining time series of average path length and the sensitivity to mutation over the course of adaptation to the sink. Figure 4*A* shows time series of populations in the source and sink habitats, figure 4*B* shows the corresponding path length, and figure 4*C* shows the sensitivity to mutation in the source and sink. Path length and sensitivity to mutation in the source generally decrease with time. Path length declines to values well below our level for truncation selection (100), presumably because mutations tend to be more deleterious in individuals with longer paths. Before adaptation in the sink, path length and sensitivity to mutation in the sink are qualitatively similar to the values in the source. (There is initially a great deal of variation in the sink because there are very few individuals there.) When adaptation does occur in the sink, however, the path length and sensitivity to mutation increase sharply in the sink population. Adaptation to the sink habitat is thus accompanied by a breakdown of canalization in the sink. As evolution in the sink habitat continues, the path length then decreases to nearly the same level as observed in the source. Continued evolution in the sink following adaptation allows canalization to reestablish. One signature of niche evolution may thus be a transient breakdown in canalization during the course of adaptation to novel harsh environments.

To explore these results further, we examined the average path length and sensitivity to mutation over multiple simulation runs in the source and compared them with those in the sink. We began by comparing the average path length and sensitivity to mutation in the source and the

Figure 4: *A*, Time series of source (*dotted line*) and sink (*solid line*) population size. *B*, Average path length of individual development in the source and sink populations for the time series in *A*. *C*, Sensitivity to mutation (mean phenotypic distance of each individual exposed to 10 different mutations) in the source and sink populations for the time series in *A*. In each panel, the dashed line indicates when migration began in the sink. Parameters are $h = 2$, $c = 0.75$, $K = 100$, and $I = 4$.

sink for four values of *c* during adaptation to the sink habitat (fig. 5*A*, 5*C*). When the sink population first reached a size of 50, indicating initial adaptation to the sink, the average path length among individuals in the sink was higher than the average path length in the source for all values of *c* (fig. 5*A*); the same was true for average sensitivity to mutation (fig. 5*C*). Furthermore, the difference between sensitivity to mutation in the source and sensitivity to mutation in the sink during adaptation was larger as the complexity of the regulation network increased. This supports the hypothesis that adaptation to the sink habitat produces a greater breakdown in canalization as the complexity of the regulation network increases.

We next compared the average path length and sensitivity to mutation in the source and sink for the same values of *c* after 10,000 generations in populations that adapted to the sink (fig. 5*B*, 5*D*). After 10,000 generations, the average path length and sensitivity to mutation in the sink were similar to the averages in the source. Thus, after adaptation to the sink habitat occurs, further evolution in the sink allows canalization to re-evolve. However, even after 10,000 generations, the average path length and sensitivity to mutation in the sink were still slightly higher

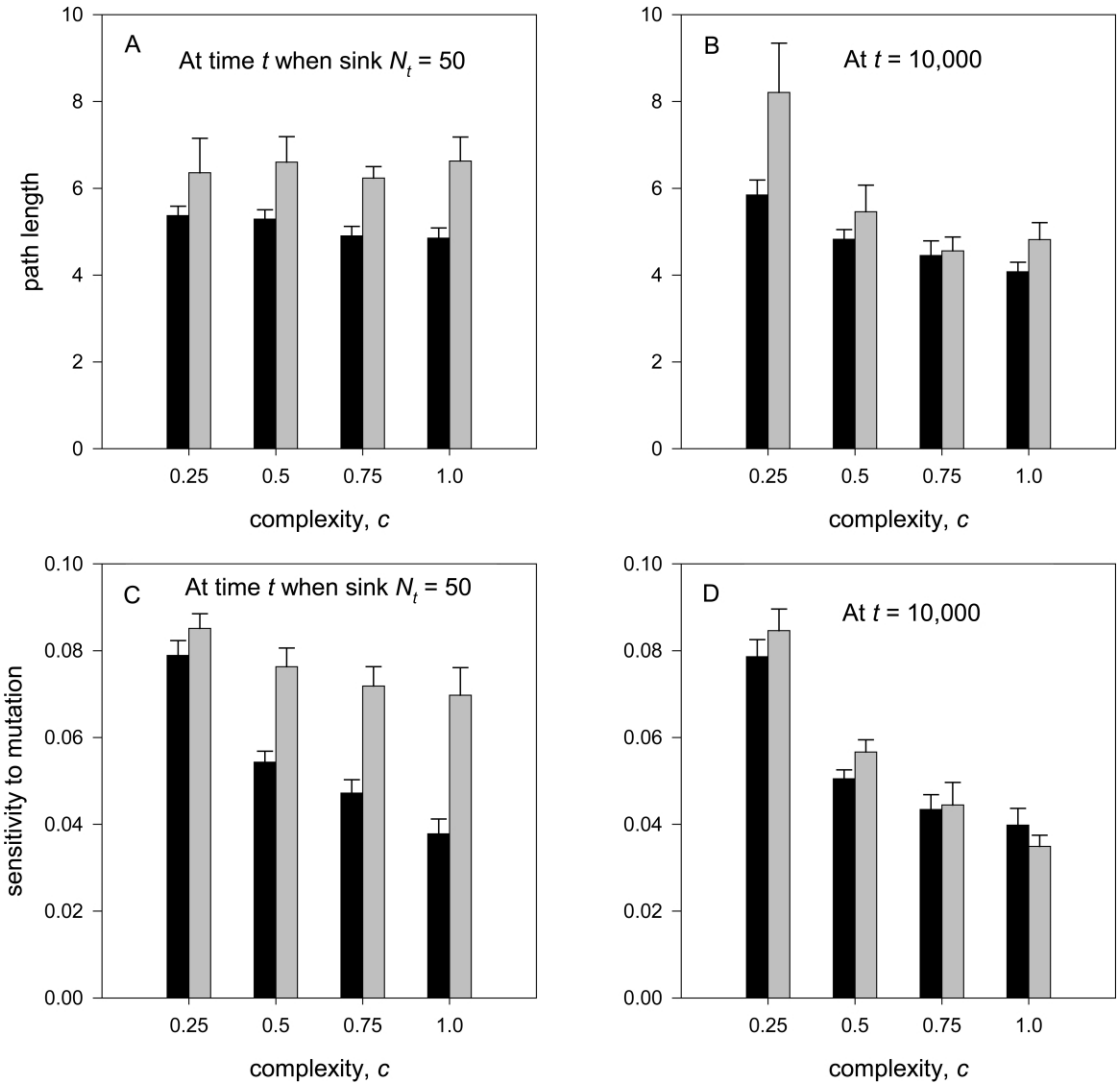


Figure 5: *A*, Path length in the source (*black bars*) and sink (*gray bars*) during adaptation (when sink population reaches 50 individuals). *B*, Path length for the same populations after 10,000 generations. *C*, Sensitivity to mutation in the source (*black bars*) and sink (*gray bars*) during adaptation to the sink habitat. *D*, Sensitivity for the same populations after 10,000 generations. The model was run 100 times for each level of complexity. In all panels, $h = 3$, $K = 100$, and $I = 4$.

than in the source for some levels of complexity, and this was especially true at the lowest level of complexity tested ($c = 0.25$).

To test whether this was simply the result of continued immigration from the source, we ran several simulations where we stopped immigration from the source when the sink population reached 50 individuals. We found that the average path length and sensitivity to mutation still tended to be larger in the sink than in the source after 10,000 generations. Consequently, a legacy of canalization breakdown may remain in the sink population long after adaptation occurs, and this may be particularly evident in less complex networks, as canalization appears to re-evolve more slowly in these networks. These same qualitative patterns were observed when we ran the simulations again with mutation rates independent of c (fig. A3).

Environmental noise acting on the initial zygote state during development had unexpected effects on the probability of adaptation in the sink. When a less complex network was examined ($c = 0.25$), both levels of environmental noise led to a higher probability of adaptation than did no noise, but low noise led to a higher probability of adaptation than did high noise (fig. 6*A*). When a more complex network was examined ($c = 0.75$), however, both levels of environmental noise still led to a higher probability of adaptation than did no noise, but the high noise level led to a higher probability than did the low noise level (fig. 6*B*). This suggests that environmental noise acting on development can increase the ability of a population to adapt to a sink habitat but that different levels of noise are better at facilitating this evolutionary transition for different complexities of the gene regulation network.

Discussion

Even though we have made very different assumptions about genetic architecture in our model, our results agreed in several respects with earlier studies of source-sink evolution that took a more classical quantitative genetic approach (e.g., Holt et al. 2003, 2004*b*, 2005*b*). We found that the sink population could stay at a small size for a long time before suddenly and rapidly increasing in number, thus creating a punctuated pattern to evolution. Moreover, increased sink harshness tended to reduce the probability of adaption to the sink habitat over a given time horizon. Similar to previous work, the probability of adapting to the sink appears to be roughly constant per unit of time, particularly at higher c (Holt et al. 2003).

We also found that increasing the number of immigrants to the sink could increase the probability of adaptation. It is instructive to compare this result to results of other recent studies that made different assumptions about genetic architecture. A single-locus model examined by Gomulkiewicz et al. (1999) showed that an increase in immigration could facilitate adaptation by increasing the pool of variation available for selection. By contrast, in quantitative genetic models of black hole sinks with fixed heritability, gene flow from the source can maintain a sink population in a stable, maladapted state (e.g., Tufto 2001; Holt et al. 2004*b*), but in the absence of density dependence at low densities, the equilibrial degree of maladaptation is independent of the rate of immigration (Holt et al. 2003). Using individual-based simulations of a quantitative trait, Holt et al. (2003) reported a positive effect of increasing immigration on the rate of adaptation to the sink and conjectured that this was due to an impact on genetic variation (as in Gomulkiewicz et al. 1999). However, subsequent analysis has shown that the individual-based model had a small Allee effect due to an assumption of monogamous mating pairs. Given Allee effects, increased immigration can increase fitness for essentially ecological reasons, which can indirectly facilitate adaptation to a sink (Holt et al. 2004*a*). When the Allee effect is removed from the individual-based quantitative genetic model, a change in the number of immigrants per generation has only a very small effect on the rate of adaptation to the sink (the effects are slightly positive with mild maladaptation in the sink and, conversely, negative with severe maladaptation; R. D. Holt and M. Barfield, unpublished results). By contrast, in this article's model, gene networks determine phenotypes. Furthermore, the individual-based model we have used has no Allee effects. Nonetheless, we found that immigration was able to increase the probability of adaptation to the sink. This suggests that the influence of immigration rate on adaptation to a sink may vary, depending on the assumptions one makes about the genetic architecture underlying the traits determining fitness.

The incorporation of a network of interacting regulatory genes into a source-sink demographic model produced several novel predictions about how adaptation may proceed in sink habitats. Whether adaptation to the sink occurs at all appears to be a function of the complexity of the "wiring" of the regulation network. Complex regulation networks lead to genetic canalization in the source habitat; this canalization breaks down in the sink during the course of adaptation to the sink habitat. More complex regulation networks are more canalized and appear less likely to undergo a breakdown in the sink habitat. Thus, complexity in genetic architecture may ultimately hamper niche evolution.

One way of testing this prediction would be to examine how a factor that decreases complexity of a regulation network affects adaptation to a sink. Gene knockout mutations occur when a gene becomes inoperative through mutation, which may thereby decrease the complexity of

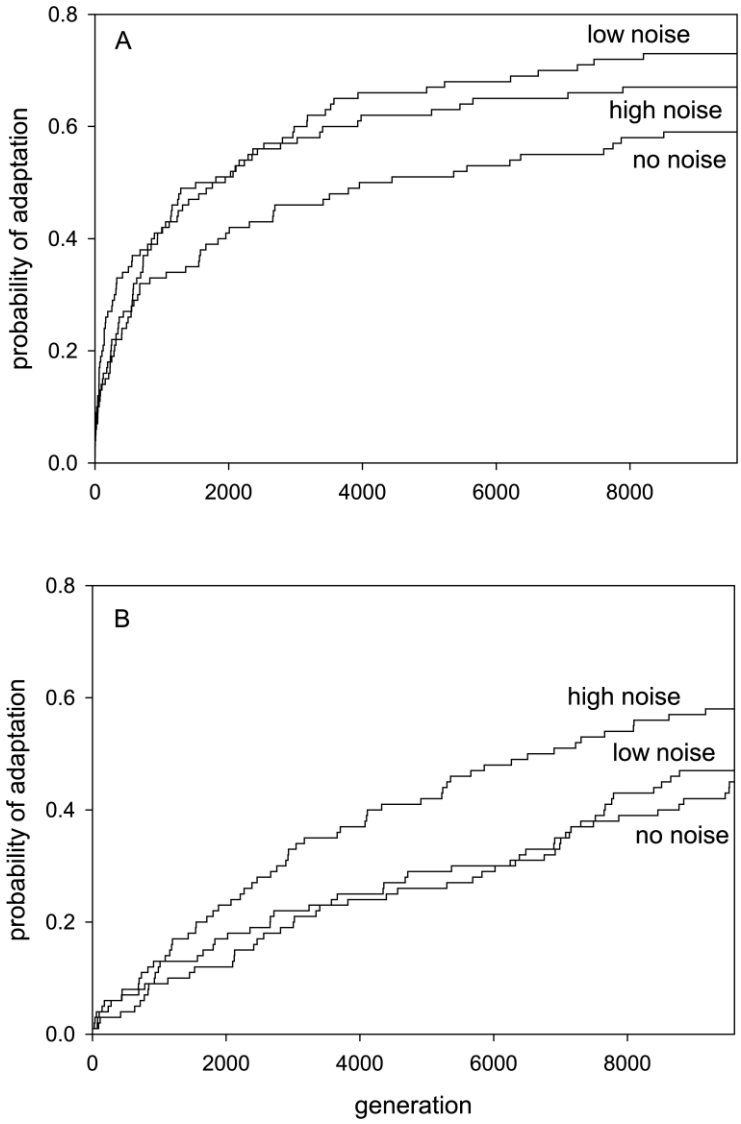


Figure 6: Probability of a population being adapted to the sink habitat as a function of time, with no developmental noise, low noise, and high noise. In *A*, $c = 0.25$; in *B*, $c = 0.75$. The model was run 100 times for each noise level in each panel. In both panels $h = 3$, $K = 100$, and $I = 4$.

the gene network. Bergman and Siegal (2003) found that yeast with gene knockout mutations showed greater variability in the expression of their other genes compared to yeast without knockout mutations. Consequently, gene knockout mutations that reduce the canalization of the regulation network may allow more rapid adaptation to a novel environment.

The interaction between an intrinsic factor (canalization) and an extrinsic factor (environmental noise) produced an unexpected outcome of our study. Holt et al. (2004*b*) found for quantitative genetic models that autocorrelated temporal variation in the quality of the sink could facilitate adaptation to sink environments. However, they observed only a weak positive effect for moderate white (uncorrelated) noise variation and a decrease in the ability to adapt to the sink with high white noise variation. In our model, noise occurred during the initial stage of development, and we found such noise increased the probability of adaptation in the sink. Furthermore, there was a relationship between the complexity of the regulation network and the level of noise that facilitated adaptation; the frequency of noise that best facilitates adaptation may

be related to *c*. If processes that create degraded habitats also increase the developmental noise that the population experiences, then those processes may also increase the probability of adaptation to the newly degraded habitats.

The ability of environmental noise to increase the probability of adaptation to the sink may be due to a process similar to genetic assimilation. Genetic assimilation occurs when an environmentally induced phenotype becomes genetically fixed by selection (i.e., canalized) and thus no longer requires the original environmental stimulus (reviewed by Pigliucci and Murren [2003]). In our model, environmental noise appears to help reveal genetic variation that can be selected on in the sink. Through selection, individuals in the sink then become canalized to the new optimal phenotype. This process occurred faster with environmental noise helping to reveal genetic variation than when environmental noise was absent.

The model of genetic canalization we use in this article represents just one approach that has been taken to modeling canalization. Some alternative models have assumed that there are noninteracting primary genes that determine the organism's phenotype and separate modifier genes that evolve to canalize the primary genes (Wagner et al. 1997; Eshel and Matessi 1998). If a single modifier gene is assumed to canalize several primary genes, then adaptation to a sink habitat may occur faster than in our model because a breakdown in canalization would require only one gene (the modifier gene) to be altered in the sink. Additionally, if genetic complexity is defined as the number of primary genes the modifier gene canalizes, then a breakdown of canalization in a population with greater genetic complexity may actually release more genetic variation and thus allow faster adaptation to the sink habitat. The effect of genetic complexity in a model with few modifier genes canalizing several primary genes may therefore be quite different from what we found for our model, where every gene had the potential to interact epistatically with every other gene.

Wagner et al. (1997) created a quite different model of canalization in which each gene that contributed to the phenotype had two variables associated with it, one determining its influence on the phenotype and a second determining the influence of the gene on the expression of other genes. A separate matrix was created establishing which genes interacted. We expect that this conceptualization of canalization in a source-sink context would produce results very similar to the results we found with our model. There would probably be a breakdown of canalization in the sink before adaptation to the sink habitat occurred. Furthermore, the probability of this breakdown would likely be lower as the matrix of gene interactions became more complex. It is less clear how environmental noise would influence adaptation in the sink, but if noise acted to increase genetic variation, then the result might be similar to our results. The implications of alternative models of canalization for evolution in heterogeneous environments are a significant challenge for future work.

All of our results have been couched in terms of source-sink dynamics because this is a scenario particularly pertinent to understanding niche evolution in heterogeneous landscapes (Holt and Gaines 1992). As mentioned in the introduction to the article, however, a sink habitat is one example among many of a novel environment to which a population is not yet adapted. Our results suggest that complexity of gene regulation networks and genetic canalization may play important roles in governing the evolutionary dimensions of many ecological phenomena. One fruitful avenue of research will be to gauge the role that gene networks and genetic canalization play in the evolution of phenotypic plasticity in novel environments (West-Eberhard 2003). A species confronted with a landscape of habitats of varying harshness may ultimately create a very complex pattern of adaptation, and species with different genetic architectures may exhibit quite different responses to the same environmental template.

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