Tuning Spatial Profiles of Selection Pressure to Modulate the Evolution of Drug Resistance

Maxwell G. De Jong¹ and Kevin B. Wood¹,²,⁺

¹Department of Physics, University of Michigan, Ann Arbor, Michigan 48109, USA
²Department of Biophysics, University of Michigan, Ann Arbor, Michigan 48109, USA

(Received 11 December 2017; published 8 June 2018)

Spatial heterogeneity plays an important role in the evolution of drug resistance. While recent studies have indicated that spatial gradients of selection pressure can accelerate resistance evolution, much less is known about evolution in more complex spatial profiles. Here we use a stochastic toy model of drug resistance to investigate how different spatial profiles of selection pressure impact the time to fixation of a resistant allele. Using mean first passage time calculations, we show that spatial heterogeneity accelerates resistance evolution when the rate of spatial migration is sufficiently large relative to mutation but slows fixation for small migration rates. Interestingly, there exists an intermediate regime—characterized by comparable rates of migration and mutation—in which the rate of fixation can be either accelerated or decelerated depending on the spatial profile, even when spatially averaged selection pressure remains constant. Finally, we demonstrate that optimal tuning of the spatial profile can dramatically slow the spread and fixation of resistant subpopulations, even in the absence of a fitness cost for resistance. Our results may lay the groundwork for optimized, spatially resolved drug dosing strategies for mitigating the effects of drug resistance.

DOI: 10.1103/PhysRevLett.120.238102

Drug resistance is a rapidly growing public health threat and a central impediment to the treatment of cancer, viruses, and microbial infections [1–4]. The battle against resistance has been largely fought at the molecular level, leading to an increasingly mature understanding of its underlying biochemical and genetic roots. At the same time, evolutionary biologists have long recognized resistance as a fundamentally stochastic process governed by the complex interplay between microbial ecology and evolutionary selection. The past decade, in particular, has seen a significant surge in efforts to develop and understand evolution-based treatment strategies to forestall resistance [5–16]. While the vast majority of this work focuses on spatially homogeneous environments, a number of recent studies, both theoretical and experimental, have demonstrated that spatial heterogeneity in drug concentration can dramatically alter the evolutionary dynamics leading to resistance [16–24].

On a practical level, the picture that emerges is somewhat bleak, as resistance evolution is dramatically accelerated in the presence of spatial gradients in drug concentration [18–20,22–24] or heterogeneous drug penetration [17,21]. Interestingly, however, recent work shows that this acceleration can be tempered or even reversed when the mutational pathway (i.e., the genotypic fitness landscape) leading to resistance contains fitness valleys [18], which are known to inhibit evolution [25–28]. Unfortunately, because the fitness landscape is a genetic property of the cells themselves, the potential for accelerated evolution appears to be “built in,” making it difficult to combat in a treatment setting. However, these results raise the question of whether non-monotonic profiles of tunable properties of the system—for example, the spatial selection pressure—might also have the potential to slow evolution, even when the mutational pathway lacks the requisite fitness valleys.

Evolution in natural or clinical settings takes place in heterogeneous environments characterized by spatial fluctuations in multiple factors, including drug concentrations, nutrients, temperature, and pH, all of which potentially affect cellular growth. Understanding evolution and ecology in such spatially extended systems is a challenging and long-studied problem [29–33]. Recent studies have demonstrated rich dynamics when intercellular interactions are defined on heterogeneous complex networks [34–36], where spatial structure can (for example) promote invasive strategies in tumor models [35] or modulate fixation times on random landscapes [34]. Remarkably, in the weak selection limit, evolutionary dynamics can be solved for any population structure [36], providing extensive insight into game-theoretic outcomes on complex networks. In addition, theoretical tools from statistical physics have proven useful for understanding spatiotemporal dynamics in spatially structured populations in a wide range of contexts, including biologically inspired Monte Carlo models [18], toy models of source-sink dynamics [19], stepping-stone models of spatial pattern formation [37], models of dispersion [38–42], and Moran metapopulation models [43–45]. In a similar spirit, here we use stochastic

DOI: 10.1103/PhysRevLett.120.238102

PHYSICAL REVIEW LETTERS 120, 238102 (2018)

0031-9007/18/120(23)/238102(6) 238102-1 © 2018 American Physical Society
models of evolution along with theoretical tools from statistical physics to investigate the effects of spatially heterogeneous fitness pressures on the evolution of resistance. In contrast to previous models defined on heterogeneous networks at the single-cell level, here we consider metapopulations connected via simple topologies and investigate the effects of spatial structure imposed by arbitrary distributions of selection pressure. While several elegant approaches exist for studying these models in particular limits (e.g., with a center manifold reduction) [43–45], here we instead use a classical mean first passage time (MFPT) approach based on adjoint equations to reduce the calculation of mean fixation times to a simple collection of linear equations that can be easily solved for arbitrary spatial distributions of selection pressures. This method also allows us to find the fixation times from arbitrary initial states, which are often difficult to compute using other methods. Using this approach, we show that resistance evolution can be either accelerated or decelerated by spatial heterogeneities in selection pressure, even when the spatially averaged selection pressure remains constant. In addition, we demonstrate that tuning the spatial distribution of selection pressure can dramatically slow fixation when the subpopulations of resistant mutants are not uniformly distributed in space.

To investigate resistance evolution on a spatially heterogeneous landscape, we consider a stochastic Moran-like model [46] of a finite population \( N \) consisting of \( (N - n^*) \) wild-type cells with fitness \( r_0 \leq 1 \) and \( n^* \) drug-resistant mutants with fitness \( r^* \), which we set to unity without loss of generality. Note that this model does not include a fitness cost to resistance (i.e., \( r^* \geq r_0 \) for all conditions). At each time step, cells are randomly selected for birth and death, with cells of higher fitness (in this case, resistant cells) chosen preferentially for division [see Supplemental Material (SM) for full model with transition rates [47]]. Wild-type cells can mutate to become drug resistant at rate \( \mu \); we neglect reverse transitions to the drug-sensitive state. To incorporate spatial heterogeneity, we consider a simple spatially extended system with \( M \) distinct microhabitats, each containing \( N \) cells; cells are allowed to migrate at rate \( \beta \) between connected microhabitats [Fig. 1(a)]. At each time \( t \), the state of the system is characterized by the vector \( n^*(x_i) \) whose components correspond to the number of mutants in each discrete microhabitat \( x_i = 0, 1, \ldots, M - 1 \). The system evolves according to a continuous time master equation,

\[
\frac{dP_m}{dt} = \sum_{m'} \Omega_{mm'} P_{m'}, \tag{1}
\]

where \( m \) and \( m' \) denote different states of the system and \( \Omega \) is a \( N^M \times N^M \) matrix whose entries depend on the wild-type fitness value \( r_0(x_i) \) at each spatial location \( x_i \) (see SM [47]). For tractability, we restrict our analysis to \( M = 3 \), which is the simplest model that allows for potentially nonmonotonic fitness landscapes, such as fitness peaks and fitness valleys. In what follows, we refer to the vector \( s(x_i) = 1 - r_0(x_i) \) as the spatial profile of selection pressure, as it measures the difference in fitness between resistant and wild-type cells in each microhabitat \( x_i \). Intuitively, large values of \( s(x_i) \) correspond to regions where the resistant mutant has a significant evolutionary advantage over the wild-type cells (e.g., regions of high drug concentration).

While Eq. (1) is difficult to solve explicitly, it is straightforward to calculate quantities that describe the evolution of resistance in various spatial profiles. The model consists of a single absorbing state—the fully resistant state \( [n^*(x_i) = N \text{ for all } x_i] \)—and the system will asymptotically approach this state. To characterize the speed of fixation in the presence of different spatial profiles \( s(x_i) \), we calculate the mean first passage times between states, which obey [48,49]
where \( T(m_j|m_i) \) is the mean time required for a system initially in state \( m_i \) to first reach state \( m_j \). We take \( m_i \) to be the fully resistant state and solve the coupled set of linear equations for \( T(m_j|m_j) \), where \( j \) is an index that runs over all initial states. In particular, when \( j \) is the fully wild-type population \( n^*(x_i) = 0 \) for all \( x_i \), we refer to the MFPT as the mean fixation time \( \tau_f \).

In the case of a single microhabitat, the mean fixation time \( \tau_f \) increases as selection pressure decreases (see SM [47]). In the spatially extended case, \( \tau_f \) would also be expected to increase when the selection pressure is globally decreased, though it should also depend on the spatial structure of the specific selection profile \( s(x_i) \). To investigate the impact of spatial structure alone, we compared \( \tau_f \) across different selection profiles \( s(x_i) \), all of which were characterized by the same spatially averaged selection pressure, \( \langle s \rangle = \sum_i s(x_i)/M \). For simplicity, we begin with a symmetric profile characterized by a background selection pressure \( s_0 \) in the edge habitats and a relative peak of height \( \delta s \) in the center habitat [Fig. 1(a)]. This toy landscape has an average selection pressure of \( \langle s \rangle = s_0 + \delta s/M \), and the parameters \( s_0 \) and \( \delta s \) are constrained by the fact that \( 0 \leq s(x_i) \leq 1 \) at all spatial locations. We vary \( \delta s \) systematically to explore different selection profiles, which can include a single selection pressure valley (\( \delta s < 0 \)), a homogeneous landscape (\( \delta s = 0 \)), or a single selection pressure peak (\( \delta s > 0 \)).

Interestingly, we find that modulating heterogeneity (\( \delta s \)) can increase or decrease \( \tau_f \) for certain choices of migration and mutation rates, even when \( \langle s \rangle \) is held constant [Fig. 1(b)]. More generally, we find that the \( \beta-\mu \) plane can be divided into three nonoverlapping regions where the homogeneous landscape (1) leads to the smallest value of \( \tau_f \), (2) leads to the largest value of \( \tau_f \), or (3) does not correspond to an extremum \( \tau_f \) [Figs. 2(a) and 2(b)]. In the latter region, heterogeneity often modulates the fixation time by only a few percent, but we do find larger effects in the high and low migration limits (i.e., on the edges) of the intermediate regime (Fig. S1 of SM [47]). In addition, as we increase \( \beta \) for a fixed value of \( \mu \), \( \tau_f \) smoothly transitions from being minimized at \( \delta s = 0 \) to being maximized near \( \delta s = 0 \) (Fig. S1 [47]). We find empirically that the fixation time can be dominated by \( \tau_1 \), the time required to achieve a small population of mutants [Fig. 2(c), rightmost panel], or \( \tau_2 \), the time required for this small population to achieve fixation [Fig. 2(c), leftmost panel]. However, in many cases—particularly those close to the intermediate region where fixation can be accelerated or slowed by heterogeneity—both timescales contribute to the dynamics. While we restrict ourselves primarily to \( N = 25 \), \( \langle s \rangle = 1/6 \), and to symmetric landscapes, we find qualitatively similar results (i.e., 3 distinct regions) for other values of \( \langle s \rangle \) (Fig. S2), \( N \) (Fig. S3), as well as for permuted selection profiles (Fig. S4), globally coupled profiles (Fig. S4), and monotonic (gradient) selection profiles (Fig. S5) (see SM [47]).
habitats achieve fixation one at a time, and fixation in a single habitat is approximated as an exponential process with rate \( \lambda(s, n_{\text{fix}}) = N(\mu + \beta n_{\text{fix}}) P_{\text{fix}}(s) \), where \( n_{\text{fix}} \) is the number of neighboring vials that have already achieved fixation and \( P_{\text{fix}} = s[1 - (1 - s)N]^{-1} \) is the probability of a single mutant fixing in a habitat with selection pressure \( s \) (see SM [47]). The approximation captures the qualitative features of fixation over a wide range of \( \mu \) and \( \beta \) (Fig. S7) and, in many cases, provides excellent quantitative agreement as well [see, for example, Fig. 2(b), left-hand panel and Fig. S7 [47]].

In general, the analytical approximation for \( \tau_f \) is algebraically cumbersome. However, in the limit \( \beta \ll \mu \), the approximation reduces to the expected maximum of three independent exponential random variables, leading to:

\[
\tau_f \approx \tau_{\text{max}} = \lambda_0^{-1} + \lambda_1^{-1} + \lambda_2^{-1} - (\lambda_0 + \lambda_1)^{-1} - (\lambda_0 + \lambda_2)^{-1} - (\lambda_1 + \lambda_2)^{-1} + (\lambda_0 + \lambda_1 + \lambda_2)^{-1},
\]

with \( \lambda_i \equiv \lambda(s(x_i), 0) \) (see SM for details [47]). In this limit, the three-vial system acts effectively as three independent systems, with the overall fixation time corresponding to the slowest fixation. After rewriting \( \tau_{\text{max}} \) in terms of \( \langle s \rangle \) and \( \delta s \), it is straightforward to show that \( \langle \partial^2 \tau_{\text{max}} / \partial \delta s^2 \rangle \big|_{\delta s = 0} < 0 \) and \( \langle \partial \tau_{\text{max}} / \partial \delta s \rangle \big|_{\delta s = 0} = 0 \), indicating that the homogeneous landscape \( (\delta s = 0) \) minimizes the fixation time, consistent with results of the exact calculation [Fig. 2(b), left-hand panel]. Intuitively, increasing heterogeneity reduces the minimum selection pressure in the spatial array, which in turn slows the expected maximum fixation time among the three habitats.

By contrast, in the limit \( \mu \ll \beta \), \( \tau_f \) reduces to the expected minimum of three independent exponential processes, leading to:

\[
\tau_f \approx \tau_{\text{min}} = (\lambda_{\text{eff}})^{-1},
\]

where \( \lambda_{\text{eff}} \equiv \lambda_0 + \lambda_1 + \lambda_2 \). In this limit, the fixation time is dominated by dynamics in the vial that first achieves fixation; the remaining vials then rapidly achieve fixation due to fast migration. For large but finite \( N \), the fixation time \( \tau_{\text{min}} \) is maximized at \( \delta s = 0 \), indicating that heterogeneity always accelerates fixation, again consistent with the exact calculation [Fig. 2(b), right-hand panel]. In this limit, the effective rate of fixation \( \lambda_{\text{eff}} \) is increased for all \( \delta \neq 0 \), as heterogeneity decreases fixation time in the vial with the fastest average fixation.

Our results indicate that a judicious choice of selection pressure profile can potentially slow fixation of \textit{de novo} mutants. In addition, selection pressure profiles can be optimized to mitigate the effects of resistance once it has emerged. One advantage of the MFPT approach [i.e., solving Eq. (2)] is that it provides fixation times starting from all possible initial states, making it straightforward to apply to cases where a resistant subpopulation already exists. Specifically, consider a situation where a resistant subpopulation has arisen at a particular spatial location. Is it possible to choose the spatial distribution of selection pressure—for example, by spatially dosing the drug—to minimize the time to fixation from this state? Intuitively, the goal is to delay the onset of treatment failure as long as possible. As an illustrative example, we consider a population consisting of \( N/2 \) mutants in the center microhabitat and calculate the mean time to fixation for different spatial profiles of selection pressure. We then find the optimal value for \( \delta s \)—that is, the heterogeneity corresponding to the spatial landscape with the slowest fixation time—in different regions of parameter space [Fig. 3(a)]. The specific choice of spatial profile significantly impacts the time to fixation from the initial resistant subpopulation [Fig. 3(b)]. We observe two distinct regions of parameter space that lead to two very different dosing regimes [Fig. 3(c)]. For \( \mu \) sufficiently large relative to \( \beta \), slowest fixation occurs when we maximize the amount of drug in the center microhabitat \( (\delta s = 0.5, \text{white region}) \). On the other hand, at large migration rates, fixation is optimally slowed by maximizing the amount of drug in the two microhabitats without any initial mutants \( (\delta s = -0.2) \).

![FIG. 3. (a) Schematic: A subpopulation of resistant mutants (red) arises at a particular spatial location. How can one choose the spatial distribution of selection pressure (i.e., drug concentration) to maximize the time to fixation? (b) Heterogeneity can significantly speed or slow fixation starting from an initial resistant subpopulation consisting of \( N/2 \) cells in the center habitat \( (\mu \equiv 10^{-9}, \beta \equiv 8 \times 10^{-3}) \). Points, exact calculation; solid line, analytical approximation. (c) The optimal spatial heterogeneity \( (\delta s) \) leading to the slowest mean fixation time from an initial state of \((0, N/2, 0)\). Depending on the specific parameter regime, the optimal selection pressure profile is the one with the largest possible valley consistent with \( \langle s \rangle \) (black) or the one having the largest possible peak (white). Red solid line, analytical approximation. (d) Relative magnitude of \( \tau_f \) (mean fixation time at maximum value of \( \delta s \)) and \( \tau_f^{\text{min}} \) (mean fixation time at minimum value of \( \delta s \)) as mutation rate decreases at constant migration rate [green arrow, (c)]. Points, exact calculation; solid line, analytical approximation. \( N = 24 \) and \( \langle s \rangle = 0.167 \) in all panels. Analytical approximation is given in Eq. (S24) of SM [47].](Image: 238102-4)
In contrast to the case with no initial mutants (e.g., Fig. 2), fixation time is never maximized by choosing the homogeneous profile. To further characterize these two regimes, we compare the fixation times from a maximally peaked landscape ($\delta s$ is maximized) to that from a landscape with a larger valley ($\delta s$ is minimized). The selection landscape that leads to the slowest fixation rapidly becomes suboptimal as mutation rate is decreased at constant $\beta$ [Fig. 3(d)].

Our model is a dramatic oversimplification of the biological dynamics leading to drug resistance. Practical applications will require analysis of more realistic models and may call for spatial optimizations with different constraints—for example, limits on the maximum allowable local selection pressure. Nevertheless, the simplicity of our model allows for a thorough characterization of fixation time over a wide range of parameters, and its behavior is surprisingly rich. Importantly, our results do not require a fitness cost of resistance or a genetic fitness valley, and they predict that spatial heterogeneity in drug concentrations would impact populations of motile and nonmotile cells in opposing ways, even when mutation rates are relatively similar. While heterogeneity is likely to accelerate evolution for populations of motile bacteria, similar to what is observed in experiments with E. coli [22,24], our results predict slow evolution for less motile cells (e.g., the nosocomial pathogen E. faecalis [50]) or cells with rapid mutation rates. Perhaps most interestingly, our results suggest counterintuitive, spatially optimal profiles for slowing the spread of resistance subpopulations. In the long term, these results may lay the groundwork for optimized, spatially resolved drug dosing strategies for mitigating the effects of drug resistance.

This work is supported by the National Science Foundation (NSF No. 1553028) and the National Institutes of Health (NIH No. 1R35GM124875-01).

kbwood@umich.edu


[27] D. M. Weinreich and L. Chao, Rapid evolutionary escape by large populations from local fitness peaks is likely in nature, Evolution 59, 1175 (2005).


[47] See Supplemental Material at http://link.aps.org/supplemental/10.1103/PhysRevLett.120.238102 which contains details of the models along with additional numerical simulations and analytical approximations.


Supplementary Information

Maxwell G. De Jong, Kevin B. Wood

1 Backward Master Equation Operator

The master equation operator $\Omega$ is populated from Markovian transition rates that come from the dynamics allowed in our system within the Moran framework. We denote $T^+[x_i]$ as a transition that increases the number of mutants in microhabitat $x_i$. Explicitly, our three transition rates increasing the number of mutants from an initial state $(j, k, \ell)$ are

\begin{equation}
T^+[x_0](j, k, \ell) = \frac{(N-j)r_0(x_0)}{(N-j)r_0(x_0) + jr^*(x_0)} \mu \\
+ \frac{jr^*(x_0)}{(N-j)r_0(x_0) + jr^*(x_0)} (1-\beta) \\
+ \frac{k\mu^*(x_1)}{(N-k)r_0(x_1) + kr^*(x_1)} \beta \tag{S1}
\end{equation}

\begin{equation}
T^+[x_1](j, k, \ell) = \frac{(N-k)r_0(x_1)}{(N-k)r_0(x_1) + kr^*(x_1)} \mu \\
+ \frac{kr^*(x_1)}{(N-k)r_0(x_1) + kr^*(x_1)} (1-2\beta) \\
+ \frac{jr^*(x_0)}{(N-j)r_0(x_0) + jr^*(x_0)} \beta \\
+ \frac{\ell\mu^*(x_2)}{(N-\ell)r_0(x_2) + \ell\mu^*(x_2)} \beta \tag{S2}
\end{equation}
and

\[
T^+[x_2](j, k, \ell) = \frac{N - \ell}{(N - \ell)r_0(x_2) + \ell r^*(x_2)}[(N - \ell)r_0(x_2)\mu + (1 - \beta)\ell r^*(x_2)] \\
+ \frac{N - \ell}{(N - k)r_0(x_1) + kr^*(x_1)}kr^*(x_1)\beta \quad (S3)
\]

Within the Moran process, we also have transitions that decrease the number of mutants in a given microhabitat. We denote \(T^-_{[x_i]}\) as the transition decreasing the number of mutants in microhabitat \(x_i\). The explicit transition rates are given by

\[
T^-[x_0](j, k, \ell) = \begin{cases} 
\frac{j}{(N - j)r_0(x_0)} & \text{Kill mutant} \\
\frac{(N - j)r_0(x_0) + j r^*(x_0)}{\text{Choose WT}} & \text{WT grows without mutation or migration} \\
\frac{(N - k)r_0(x_1)}{\text{Kill mutant}} & \text{Pick WT} \\
\frac{(N - k)r_0(x_1) + kr^*(x_1)}{\text{Right migration}} & \beta
\end{cases} \quad (S4)
\]

\[
T^-[x_1](j, k, \ell) = \begin{cases} 
\frac{k}{(N - k)r_0(x_1)} & \text{Kill mutant} \\
\frac{(N - k)r_0(x_1) + kr^*(x_1)}{\text{Choose WT}} & \text{WT grows without mutation or migration} \\
\frac{(N - j)r_0(x_0)}{\text{Kill mutant}} & \text{Pick WT} \\
\frac{(N - j)r_0(x_0) + j r^*(x_0)}{\text{Left migration}} & \beta \\
\frac{(N - \ell)r_0(x_2)}{\text{Kill mutant}} & \text{Pick WT} \\
\frac{(N - \ell)r_0(x_2) + \ell r^*(x_2)}{\text{Right migration}} & \beta \quad (S5)
\end{cases}
\]

and

\[
T^-[x_2](j, k, \ell) = \begin{cases} 
\frac{\ell}{(N - \ell)r_0(x_2)} & \text{Kill mutant} \\
\frac{(N - \ell)r_0(x_2) + \ell r^*(x_2)}{\text{Choose WT}} & \text{WT grows without mutation or migration} \\
\frac{(N - k)r_0(x_1)}{\text{Kill mutant}} & \text{Pick WT} \\
\frac{(N - k)r_0(x_1) + kr^*(x_1)}{\text{Left migration}} & \beta \quad (S6)
\end{cases}
\]

With these transition rates, \(\Omega\) is populated according to

\[
\Omega_{m,n} = \begin{cases} 
W^{n\rightarrow m} & n \neq m \\
- \sum_{m' \neq m} W^{n\rightarrow m'} & n = m \quad (S7)
\end{cases}
\]
where $W_{n \rightarrow m}$ is the transition rate from initial state $n$ to final state $m$. Note that the number of mutants can change by one at most, so the backwards master equation operator is sparse.

We can then solve the mean first passage time equation

$$-1 = \sum_{m' \neq m_f} T(m_f|m')\Omega_{n',m_i}$$

where $T(m_f|m_i)$ is the mean time required for a system initially in state $m_i$ to first reach state $m_f$. All of the natural boundary conditions are already imposed by $\Omega$, but we do need to specify that

$$T((n^*(x_0), n^*(x_1), n^*(x_2))|(n^*(x_0), n^*(x_1), n^*(x_2))) = 0$$

for any initial state $(n^*(x_0), n^*(x_1), n^*(x_2))$, which is intuitively obvious.

2 Characterization of Intermediate Regime

To further investigate evolution in the intermediate regime, where heterogeneity can either speed or slow fixation, depending on the specific profile, we calculated the minimum and maximum fixation times ($\tau_{min}^f$ and $\tau_{max}^f$, respectively) as $\delta s$ is modulated to create different selection pressure distributions (Fig. S1a). While in many cases the fixation time is decreased by only a few percent, we do find larger effects in the high and low migration limits (i.e. on the edges) of the intermediate regime. To understand how the spatial $\tau_f$ profiles change over this intermediate region, we fix $\mu$ and traverse across a trajectory in $\beta$ as shown by the arrow in Fig. S1a. As we increase $\beta$, $\tau_f$ smoothly transitions from being minimized at $\delta s = 0$ to being maximized near $\delta s = 0$ (Fig. S1b).

3 Alternative Landscape Parameters

The results in the main text were all generated with $\langle s \rangle = 0.167$. However, the existence of three regions of parameter space is not specific to this particular value of $\langle s \rangle$. In Fig. S2, we see that we observe three regions of parameter space with a variety of values for $\langle s \rangle$. It is worth noting that the relative size and location of the intermediate region change with $\langle s \rangle$, and the magnitude of effects also changes with $\langle s \rangle$. However, the existence of these regions is constant throughout these different classes of landscapes.

Note that a second intermediate region appears with larger values of $\langle s \rangle$. The existence of this additional intermediate region does not follow the same intuitive understanding and is more subtle. This second intermediate region arises due to numerical reasons. Upon discretizing our selection landscape in steps of $\Delta \delta s = 0.1$, it is possible for the “true” minimum to occur near $\delta s = 0$ but not exactly at this value. With these relatively large step sizes, it allows the “true” minimum to be mapped to $\delta s = 0$ even though the true minimum
Figure S1: (a) Minimum fixation times ($\tau^\text{min}_f$) over different selection pressure distributions (relative to the maximum fixation times, $\tau^\text{max}_f$) in the intermediate parameter region where fixation can be both accelerated and decelerated. $N = 25$ and $\langle s \rangle = 0.167$. (b) Across a specific trajectory in the intermediate region (arrow in panel (a)), the dependence of $\tau_f$ on heterogeneity ($\delta s$) transitions smoothly from a state with a minimum at $\delta s = 0$ (light test curve) to one with a maximum at $\delta s = 0$ (darkest curve). For ease of comparison, fixation times are scaled to arbitrary units between 0 and 1.

occurs at some small non-zero value. So the apparent area between the two intermediate regions is actually an artifact of the numerical discretization of our landscape. Further evidence comes from looking at the fixation times in this second intermediate region, which differ by less 1% from the spatially homogeneous landscape, so it is likely of little significance.

We can also vary $N$ to confirm that the existence of the three regions of parameter space is resilient to changes in $N$. We investigated a few different values of $N$ in Fig. S3, and we again see the same three regions of parameter space.

4 Alternative Topologies

We can also confirm that the existence of three regions of parameter space is not predicated upon the specific topology chosen. The results of the main text assumed a nearest-neighbor connection without migration between the boundary microhabitats. We can easily imagine additional topologies for a specified selection landscape. We can remove one of the existing connections and allow migration between the boundary microhabitats (which is equivalent to cyclically permuting the selection landscape with the same connections). The resulting phase plot is shown in Fig. 4(a). We can also imagine a global topology in which all microhabitats are connected to all others. The phase plot from this global topology is shown in Fig. 4(b). Note that the phase plots differ quantitatively when the topology is changed, but all three topologies support three different
Figure S2: Changing $\langle s \rangle$ does not change the existence of three regions of our parameter space. The appearance of a second intermediate region is artificial and due to numerical discretization.

Figure S3: Different population sizes $N$ result in the same qualitative behavior.
Figure S4: Different topologies also produce three unique regions of parameter space.

Figure S5: A monotonic selection landscape produces a qualitatively similar phase plot to that generated with a non-monotonic landscape.

regions of parameter space.

5 Monotonic Landscape

We can also imagine using a monotonic selection landscape. A non-monotonic landscape potentially allows for more complicated dynamics due to the lack of a single direction of selection pressure gradient. In Fig. S5, we see that a monotonic landscape also results in three regions of parameter space. Future work will be focused on understanding the role of monotonicity on the fixation times.
6 Analytical Approximation for Single Habitat

In a single microhabitat with selection pressure $s$, we can approximate the fixation time $\tau_f(s)$ in the limit $\mu \ll 1$ as

$$\tau_f(s) \approx \frac{1}{\lambda_{eff}} = \frac{1}{\mu N P_{fix}(s)}.$$  (S10)

Equation S10 provides a good approximation to $\tau_f(s)$ for a range of $\mu \ll 1$ (Figure S6). This approximation assumes that mutations are sufficiently rare that fixation times are dominated by arrival and fixation of a single mutant (i.e. the timescale of fixation of a single mutant is fast compared to the expected arrival time of the next mutant via mutation). Therefore, the arrival time distribution is exponential with a rate parameter $\lambda_{eff}$. The factor $\mu N$ is the transition rate $T^+(0)$ for increasing the number of mutants by one starting from zero mutants (see, for example, Equation S1 with $j = 0$ and $\beta = 0$). $P_{fix}$ is the probability of fixation of a single mutant in a habitat with selection pressure $s$ and no mutation. The expression for $P_{fix}(s)$ is well-known, but for completeness, we briefly repeat the derivation here. To do so, we first write an iterative equation for $p_i$, the probability of fixation from a starting condition of $i$ mutants. We have

$$p_i = T^-(i) p_{i-1} + T^+(i) p_{i+1} + (1 - T^+(i) - T^-(i)) p_i$$

$$= \frac{\rho}{1 + \rho} p_{i-1} + \frac{1}{1 + \rho} p_{i+1}.$$  (S11)

where $\rho \equiv T^-(i)/T^+(i) = 1 - s$, and we have used the fact that

$$T^+(i) = \frac{i(N - i)}{N - s(N - i)}$$
$$T^-(i) = \frac{i(N - i)(1 - s)}{N - s(N - i)}.$$  (S12)

Equation S11 is a second order linear difference equation with constant coefficients. It has general solution $p_i = c_1 + c_2 \rho^i$, and given the boundary conditions $p_0 = 0$ and $p_N = 1$, we can solve for the constants $c_1$ and $c_2$ to arrive at

$$p_i = \frac{1 - \rho^i}{1 - \rho^N}.$$  (S13)

For $i = 1$ and $\rho = 1 - s$, we therefore have

$$P_{fix}(s) \equiv p_1 = \frac{s}{1 - (1 - s)^N}.$$  (S14)

7 Analytical Approximation for Multiple Habitats

To derive an approximate expression for $\tau_f$ in the $M = 3$ vial array, we again consider the limit where the arrival of the first mutant in each vial dominates
Within a single vial system, the fixation time is well-approximated for sufficiently small $\mu$ by Equation S10. Circles: exact calculation; solid line: approximation. Mutation rates are $\mu = 10^{-7}$ (blue), $\mu = 10^{-5}$ (red), and $\mu = 10^{-3}$ (black). $N = 75$ for all curves. Fixation times are measured in units of $N^{-1}$. 
the fixation time, which now corresponds to $\mu, \beta \ll 1$. In this limit, the time to fixation within a single vial—given the presence of a single initial mutant—is again fast compared to the arrival time of that first mutant, which now can be due to either a mutation event (at rate $\mu N$) or a migration event (at rate $N \beta n_{fix}$, where $n_{fix}$ is the integer number of (connected) neighboring vials that have already achieved fixation). Therefore, the different habitats effectively achieve fixation one at a time, and the fixation time within each vial is an exponential process governed by a rate

$$\lambda(s, n_{fix}) = N(\mu + \beta n_{fix})P_{fix}(s)$$

(S15)

with $P_{fix}(s)$ again given by Equation S14. For economy of notation, we introduce the shorthand $\lambda_i(n_{fix}) \equiv \lambda(s(x_i), n_{fix})$. The global fixation time, which depends on the distribution of selection pressures in the three habitats, can be approximated by

$$\tau_f \approx \tau_{min} + Q(x_0)\tau_{max}^\beta(x_1, x_2) + Q(x_1)\tau_{max}^\beta(x_0, x_2) + Q(x_2)\tau_{max}^\beta(x_0, x_1).$$

(S16)

$\tau_{min}$ is the fixation time of the fastest vial in the absence of migration and is given by the expected minimum of three independent, exponentially distributed variables with rates $\lambda_i(0)$,

$$\tau_{min} = \frac{1}{\lambda_0(0) + \lambda_1(0) + \lambda_2(0)}. \quad \text{(S17)}$$

$Q(x_i)$ is the probability that the vial at location $x_i$ was the first to reach fixation in the absence of migration, and it is given by

$$Q(x_i) = \frac{\lambda_i(0)}{\sum_{j=0}^{2} \lambda_j(0)} \quad \text{(S18)}$$

The $\beta$ dependence of Equation S16 is contained in the terms $\tau_{max}^\beta(x_i, x_j)$, which also implicitly contain information about the connection topology. $\tau_{max}^\beta(x_i, x_j)$ is the expected maximum time of two independent fixation events occurring in habitats $x_i$ and $x_j$, assuming that the third habitat ($x_k \neq x_i, x_j$) has already achieved fixation and can therefore supply seed mutants at a rate $\beta N$. For example, we have

$$\tau_{max}^\beta(x_0, x_1) = \frac{1}{\lambda_0(0) + \lambda_1(1)} + \frac{\lambda_0(0)}{\lambda_0(0) + \lambda_1(1)} \left(\lambda_1(2)\right)^{-1} + \frac{\lambda_1(1)}{\lambda_0(0) + \lambda_1(1)} \left(\lambda_0(1)\right)^{-1}$$

(S19)

where the first term is the expected minimum time of two independent processes occurring at rates $\lambda_0(0)$ and $\lambda_1(1)$; note that since the vial at $x_2$ has already achieved fixation, the rate for the vial at $x_1$ is taken at $n_{fix} = 1$, which accounts for migration from vial $x_2$ to $x_1$. The second term in Equation S19 is a product of two terms: the probability that the vial at $x_0$ achieves fixation before the vial at $x_1$, and the rate at which the vial at $x_1$ would achieve fixation ($1/\lambda_1(2)$). The latter rate corresponds to $n_{fix} = 2$, as vials at $x_0$ and $x_2$ have already
achieved fixation and can both contribute $\beta N$ to the arrival rate of the first mutant. Finally, the last term is a product of two terms: the probability that the vial at $x_1$ achieves fixation before the vial at $x_0$, and the rate at which the vial at $x_0$ would achieve fixation ($1/\lambda_0(1)$).

Because of the symmetry of the selection pressure profiles considered here ($s(x_0) = s(x_2)$), we have $\tau_{\text{max}}^\beta(x_0,x_2) = \tau_{\text{max}}^\beta(x_1,x_2)$, while the remaining term in Equation S16 is given by

$$\tau_{\text{max}}^\beta(x_0,x_2) = \frac{1}{\lambda_0(1) + \lambda_2(1)} + \frac{\lambda_0(1)}{\lambda_0(1) + \lambda_2(1)}(\lambda_2(1))^{-1} + \frac{\lambda_2(1)}{\lambda_0(1) + \lambda_2(1)}(\lambda_0(1))^{-1}$$

where the last line accounts for the symmetry of the selection profile.

By combining Equation S16 with Equations S14, S15, and S17-S20, we arrive at a general equation for $\tau_f$ that depends on the selection pressure profile, $\mu$, $\beta$, and $N$. The general expression is long and cumbersome, but it is straightforward to numerically evaluate $\tau_f$ for any value of the parameters. We find that the approximation performs surprisingly well over a large range of parameter values, qualitatively reproducing all features of the full MFPT calculation (Figure S7, main panel; Compare to Figure 2, main text) and even providing excellent quantitative agreement in many cases (Figure S7, bottom panels). To gain additional analytical insight into the model, we consider in what follows several limiting cases where additional analytical progress is tractable.

### 7.1 $\beta \ll \mu$ Limit

To investigate the limit $\beta \ll \mu$, we expand Equation S16 in the small parameter $\epsilon_1 \equiv \beta/\mu$ and neglect terms of order $\epsilon_1$ and higher. In this limit, $\tau_f$ in Equation S16 reduces to $\tau_{\text{max}}^\beta$, given by

$$\tau_{\text{max}}^\beta = \frac{2}{\lambda_0(0)} + \frac{1}{\lambda_1(0)} - \frac{2}{\lambda_0(0) + \lambda_1(0)} - \frac{1}{\lambda_0(0) + \lambda_2(0)} + \frac{1}{2\lambda_0(0) + \lambda_1(0)}$$

which is the expected maximum of three independent exponentially distributed random variables; note that we have again taken into account the symmetry in the selection pressure profile ($\lambda_0 = \lambda_2$). In this limit, the three vial system acts effectively as three independent systems, with the overall fixation time corresponding to the slowest fixation. After rewriting $\tau_{\text{max}}$ in terms of $\langle s \rangle$ and $\delta s$, it is straightforward (though algebraically tedious) to show that $\partial \tau_{\text{max}}/\partial \delta s|_{\delta s=0} = 0$ and $\partial^2 \tau_{\text{max}}/\partial (\delta s)^2|_{\delta s=0} > 0$, indicating that the homogeneous landscape ($\delta s = 0$) minimizes the fixation time, consistent with results of the exact calculation. Intuitively, increasing heterogeneity reduces the minimum selection pressure in the spatial array, which in turn slows the expected maximum fixation time among the three habitats.
Figure S7: Parameter regimes and fixation time curves calculated using approximate MFPT (compare to Figure 2, main text). MFPT approximations were performed for the indicated values of $\beta$ and $\mu$ and for $-0.2 \leq \delta s \leq 0.5$ in steps of 0.025. Red numbers on the main plot correspond to curves shown in the subplots below. Black circles: exact calculation. Red curves: approximation.
Figure S8: Fixation times near the origin as a function of $\delta s$ in the $N \to \infty$ limit. $\beta$ and $\mu$ values for panels labeled 1-10 are the same as in Figure S7; we’ve also included $\beta = 0$ and $\beta = 1$. $\langle s \rangle = 1/6$ (red) and $\langle s \rangle = 5/6$ (blue).

### 7.2 $\beta \gg \mu$ Limit

To investigate the limit $\beta \gg \mu$, we expand Equation S16 in the small parameter $\epsilon_2 \equiv \mu/\beta$. The dominant term is of order $1/\epsilon_2$, and we ignore terms of order unity and higher. In the limit $\beta \gg \mu$, $\tau_f$ in Equation S16 reduces to the expected minimum of three independent exponential processes, leading to $\tau_f \approx \tau_{\text{min}}$ (Equation S17). In this limit, the fixation time is dominated by dynamics in the vial that first achieves fixation; the remaining vials then rapidly achieve fixation due to fast migration. For large but finite $N$, the fixation time $\tau_{\text{min}}$ is maximized at $\delta s = 0$, indicating that heterogeneity always accelerates fixation, again consistent with the exact calculation (Fig. 2b, right panel). In this limit, the effective rate of fixation $\lambda_{\text{eff}}$ is increased for all $\delta \neq 0$, as heterogeneity decreases fixation time in the vial with the fastest average fixation.

### 7.3 Large $N$ Limit

While our primary focus is on finite populations, we briefly consider here the limit of $N \to \infty$, where Equation S16 reduces (after simplification) to

$$\tau_f \approx \frac{1}{\mu(s_0 + s_1)} \left[ 1 + \frac{3\mu s_1}{2(\beta + \mu)s_0} + \frac{2\mu (2s_1(s_0 + s_1) + \mu(s_0^2 + s_0s_1 + s_1^2))}{(2\beta + \mu)s_1(\beta s_1 + \mu(s_0 + s_1))} \right]$$  \hspace{1cm} (S22)

Note that prior to taking the $N \to \infty$ limit, we first rescaled our time units by a factor of $N$. We’ve also used the shorthand $s_i \equiv s(x_i)$ and consider only symmetric landscapes ($s_0 = s_2$).

In the limit $s_1/s_0 \to 0$ (large selection valley), Equation S22 reduces to $\tau_f^\infty = 1/(s_1(2\beta + \mu))$, which corresponds to selection in a single vial of selection pressure $s_1$ where seed mutants can arise due to mutation ($\mu$) or due to migration.
from one of two vials that have already achieved fixation \((2\beta)\). Fixation in the center vial is the rate limiting step for global fixation. Similarly, when \(s_0/s_1 \to 0\) (large selection peak), we have \(\tau_F^\infty = 3/(2s_0(\beta + \mu))\), which is the maximum of two independent exponential processes that each occur at rate \(s_0(\beta + \mu)\). Fixation in the peripheral vials is the rate-limiting process.

To evaluate the magnitude of the change in fixation time as the selection profile is modulated, we fix the average selection pressure \(\langle s \rangle\) and calculate the difference in fixation times \(\Delta \tau\) between a selection profile with a large peak \((s_0 = s_2 \sim \epsilon_0)\) and one with a large valley \((s_1 \sim 2\epsilon_0)\), where \(\epsilon_0 \ll 1\) is assumed to be small so that the vast majority of the selection pressure is concentrated either in the center vial or the edge vials. By expanding Equation S22 in \(\epsilon_0\) and ignoring terms of order \(\epsilon_0\) or higher, we have

\[
\frac{\Delta \tau}{\tau_v} = \frac{\tau_p - \tau_v}{\tau_v} = \frac{5 + 2\gamma}{1 + \gamma} > 0,
\]

where \(\tau_v\) (\(\tau_p\)) corresponds to the selection profile with a large valley (peak) and \(\gamma \equiv \mu/\beta\). The fixation time is always larger (slower) in a sharply peaked landscape than one with a sharp valley. Choosing the peaked landscape over the valley leads to a relative increase of 2-5 fold depending on the ratio of mutation to migration.

Following a change of variables in Equation S22 from \(\{s_i\} \to \langle s \rangle\) and \(\delta s\), we find that the homogeneous landscape is an optimum \((\partial \tau_F^\infty / \partial \delta s = 0)\) only for \(\beta = 0\) and \(\beta = \mu \left(7 + \sqrt{145}\right)/4\), and each corresponds to a minimum of the fixation time. Surprisingly, then, all other values of \(\beta\) correspond to the intermediate regime, where heterogeneity can either slow or speed fixation depending on the specific landscape. In practice, the value of \(\delta s\) for which a minimum occurs is often very close to zero, though it does depend on \(\langle s \rangle\) (Figure S8). Large heterogeneity–either a peak \((\delta s > 0)\) or a valley \((\delta s < 0)\)–tends to slow fixation. For \(\beta \gg \mu\), the fixation time reduces to \(\tau_F^\infty \approx (3\mu(s))^{-1}\) and no longer depends on \(\delta s\); this corresponds to the fixation time of a single habitat with effective selection pressure \(s_e = 3\langle s \rangle\), where the factor of 3 results from a tripling of the population size (relative to a single vial).

### 7.4 Time to Fixation With Initial Mutants

We now consider the case when there are \(N/2\) resistant mutants in the middle habitat \((x_1)\). In this case, we can approximate the time to fixation \(\tau_F^{N/2}\) as

\[
\tau_F^{N/2} \approx [P_{fix}(s_1, N/2)]\tau_{max}^\beta(x_0, x_2) + (1 - P_{fix}(s_1, N/2))\tau_f
\]

where \(\tau_{max}^\beta(x_0, x_2)\) is given by Equation S20, \(\tau_f\) is the fixation time starting from zero initial mutants (Equation S16), and \(P_{fix}(s, N/2)\) is the probability of fixation in a single vial with selection pressure \(s\) starting from \(N/2\) initial mutants,

\[
P_{fix}(s, N/2) = \frac{1 - (1 - s)^{N/2}}{1 - (1 - s)^N}.
\]
The first term in Equation S24 corresponds to the middle vial achieving fixation first, while the second term corresponds to another vial first achieving fixation. As before, we are assuming sufficiently small $\mu$ and $\beta$ that the fixation (or extinction) of a mutant population in a single vial happens much faster than the typical arrival time of a new mutant via migration or mutation. In the limit $N \to \infty$, the middle vial is guaranteed to reach fixation first, and Equation S24 reduces to $\tau_f^{N/2} = 3/(2s_0(\beta + \mu))$, which is the expected maximum of the (independent) fixation times for the edge vials. In this limit, it is always optimal to distribute the drug in the habitats with no initial mutants (i.e. minimize $s_0$).

For general $N$, fixation in the middle habitat is not guaranteed, and the optimal strategy will depend on $\mu$ and $\beta$. While Equation S24 is a cumbersome expression that depends nonlinearly on the system parameters, it is easy to evaluate numerically, and we find that it provides an excellent approximation to the exact results in the parameter regimes considered here (Figure 3b-d).