Equilibrium Distribution of Mutators in the Single Fitness Peak Model

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This Letter develops an analytically tractable model for determining the equilibrium distribution of mismatch repair deficient strains in unicellular populations. The approach is based on the single fitness peak model, which has been used in Eigen's quasispecies equations in order to understand various aspects of evolutionary dynamics. As with the quasispecies model, our model for mutator-nonmutator equilibrium undergoes a phase transition in the limit of infinite sequence length. This "repair catastrophe" occurs at a critical repair error probability of $\epsilon_r = L_{\rm via}/L$, where $L_{\rm via}$ denotes the length of the genome controlling viability, while L denotes the overall length of the genome. The repair catastrophe therefore occurs when the repair error probability exceeds the fraction of deleterious mutations. Our model also gives a quantitative estimate for the equilibrium fraction of mutators in *Escherichia coli.*

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In order to preserve the integrity of their genomes, living systems have evolved sophisticated mechanisms for correcting errors in their DNA sequences [1]. Otherwise, genetic damage due to environmental factors such as radiation, metabolic free radicals, and mutagens, combined with replication errors, would lead to unviable organisms due to the unrecoverable loss of genetic information. This phenomenon, which was first characterized by Eigen in [2], is known as the "error catastrophe." It has since been studied in a number of theoretical papers [3–5] (and references therein) and has also been observed experimentally [6].

Some of the error-correcting ability in living systems is already built into the DNA polymerases themselves. In *Escherichia coli*, the proofreading ability of the DNA replicase Pol III results in an error probability of 10^{-7}–10^{-6} per base pair [1]. Additional enzymes continuously scan the DNA molecule, repairing lesions and mismatches that occur due to environmental damage.

A key error-repair mechanism is known as mismatch repair and occurs immediately following DNA replication. The mismatch repair system scans the DNA molecule, identifies, and then corrects mismatched base pairs. Mismatch repair in *E. coli* reduces the error probability in DNA replication to 10^{-10}–10^{-8} per base pair [1]. Cells with inactivated mismatch repair consequently have mutation rates which are 10 to 10 000 times higher than cells whose mismatch repair system is functioning. Because of their higher than wild-type mutation rates, these "mutator" strains are believed to play an important role in the emergence of antibiotic resistance, and cancer in multicellular organisms [7–12].

To develop a model for the equilibrium distribution of mutators versus nonmutators in a unicellular population, we consider a genome of alphabet size S ("bases" $0, 1, \ldots, S-1$), consisting of two genes. The first gene consists of $L_{\rm via}$ bases and controls the viability of the genome. The second gene consists of $L_{\rm rep}$ bases and codes for the enzymatic machinery involved in mismatch repair. If we let σ denote an arbitrary gene sequence, then we may write $\sigma = \sigma_{\rm via}\sigma_{\rm rep}$.

We assume a single fitness peak (SFP) model for both genes. Thus, there is a unique "fit" sequence $\sigma_0 = \sigma_{\rm via,0} \times \sigma_{\rm rep,0}$. A cell with genome σ has a first-order growth rate constant $k \gg 1$ if $\sigma_{\rm via} = \sigma_{\rm via,0}$, and 1 otherwise. Mismatch repair has an error probability of ϵ_r per mismatched base pair and is functioning only when $\sigma_{\rm rep} = \sigma_{\rm rep,0}$.

While somewhat artificial, the SFP model has been successfully applied in [13,14] toward understanding the correlations between antibody and viral mutation rates. Furthermore, because proteins generally have a key set of conserved residues which more or less dictate their final structure and function, the corresponding gene has a subsequence of conserved bases required for its proper function [1,15]. Thus, by summing over the unconserved bases, it is possible to reduce the fitness landscape to an SFP in the conserved subsequence. Therefore, there is reason to believe that many of the phenomenological aspects of our system can be captured by an SFP-based approach and that such an approach can also be semiquantitative in a number of cases.

The basic equation governing the dynamics on the genome space has the form of Eigen's quasispecies equations [2],

$$\frac{dx_\sigma}{dt} = [\kappa_\sigma - \bar{\kappa}(t)]x_\sigma + \sum_{\sigma' \neq \sigma} [\kappa_m(\sigma', \sigma)x_{\sigma'} - \kappa_m(\sigma, \sigma')x_\sigma], \quad (1)$$

where x_σ denotes the fraction of the population with genome σ, κ_σ is the growth rate constant of σ, $\kappa_m(\sigma, \sigma')$ denotes the mutation rate constant from σ to σ', and $\bar{\kappa}(t) \equiv \sum_\sigma \kappa_\sigma x_\sigma(t)$.

We assume that replication errors are sufficiently small so that we need worry only about point mutations, and that since $k \gg 1$, any flow off of the viability peak is unidirectional. Furthermore, since we are interested only in the relative distribution of viable mutators and

nonmutators, we focus only on the "repair" subspace of sequences given by $\sigma = \sigma_{\text{via},0}\sigma_{\text{rep}}$. Note that we are assuming that the system is well below the error threshold. The reasoning behind this assumption is that mutators, despite their higher-than-wild-type mutation rates, are still viable organisms and therefore live well below the error catastrophe. This also allows us to treat the physics of mutator/nonmutator equilibrium separately from the physics of the classical error threshold. We leave the incorporation of the error catastrophe into this model for future work.

From now on, we simplify matters and redenote σ_{rep} as σ. The full genome is $\sigma_{\text{via},0}\sigma$ by implication. On this subspace, the effective growth rate constant becomes $k(1 - L_{\text{via}}\epsilon_\sigma)$, due to leakage off of the fitness peak. Here ϵ_σ denotes the per base pair replication error probability and is equal to $\epsilon\epsilon_r$ if $\sigma = \sigma_0$, and ϵ otherwise. Finally, by the symmetry of our system, we may make the further assumption that x_σ depends only on the Hamming distance $\text{HD}(\sigma, \sigma_0)$ from σ_0. Thus, defining $\Omega_l(\sigma) = \{\sigma' | \text{HD}(\sigma', \sigma) = l\}$, we may then also define $x_l = x_\sigma$, where $\sigma \in \Omega_l(\sigma_0)$. Note that point mutations between x_l and some x_σ may occur only if $x_\sigma \in \Omega_{l,l\pm1}(\sigma_0)$. However, we may negect intra-$\Omega_l(\sigma_0)$ couplings, due to cancellation of mutational inflows and outflows.

A $\sigma \in \Omega_l(\sigma_0)$ may be connected via a point mutation to a $\sigma' \in \Omega_{l-1}(\sigma_0)$ by changing any one of the l bases distinct from the corresponding bases in σ_0 back to the corresponding base in σ_0. Thus, there are l possible connections. A $\sigma \in \Omega_l(\sigma_0)$ may be connected via a point mutation to a $\sigma' \in \Omega_{l+1}(\sigma_0)$ by changing any one of the $L_{\text{rep}} - l$ bases equal to the corresponding bases in σ_0. Since there are $S - 1$ possibilities per base, the result is $(L_{\text{rep}} - l)(S - 1)$ connections. The net mutational flow is then

$$\sum_{\sigma' \neq \sigma} [\kappa_m(\sigma', \sigma)x_{\sigma'} - \kappa_m(\sigma, \sigma')x_\sigma] = \frac{kl}{S-1}(\epsilon_{l-1}x_{l-1} - \epsilon_l x_l) + k(L_{\text{rep}} - l)(\epsilon_{l+1}x_{l+1} - \epsilon_l x_l), \tag{2}$$

where $\epsilon_0 = \epsilon\epsilon_r$, and $\epsilon_l = \epsilon$ for $l \geq 1$. We divide the ϵ's by $S - 1$ because a point mutation can occur to any one of the $S - 1$ bases distinct from the changed base.

We also have $\kappa(t) = k(1 - L_{\text{via}}\epsilon) + kL_{\text{via}}\epsilon(1 - \epsilon_r)x_0$. Now, define $C_l = \binom{L_{\text{rep}}}{l}(S-1)^l$, the number of elements in $\Omega_l(\sigma_0)$, and set $z_l = C_l x_l$. If we reexpress the dynamical equations in terms of z_l, then at equilibrium we obtain the system of equations

$$\begin{aligned}
0 &= \frac{L_{\text{via}}}{L_{\text{rep}}}(1 - \epsilon_r)z_0(1 - z_0) + \frac{z_1}{L_{\text{rep}}(S-1)} - \epsilon_r z_0, \\
0 &= -\frac{L_{\text{via}}}{L_{\text{rep}}}(1 - \epsilon_r)z_0 z_1 + \epsilon_r z_0 - \left(1 - \frac{1}{L_{\text{rep}}} + \frac{1}{L_{\text{rep}}(S-1)}\right)z_1 + \frac{2}{L_{\text{rep}}(S-1)}z_2, \\
&\vdots \\
0 &= -\frac{L_{\text{via}}}{L_{\text{rep}}}(1 - \epsilon_r)z_0 z_l + \left(1 + \frac{1}{L_{\text{rep}}} - \frac{l}{L_{\text{rep}}}\right)z_{l-1} - \left(1 - \frac{l}{L_{\text{rep}}} + \frac{l}{L_{\text{rep}}(S-1)}\right)z_l + \frac{l+1}{L_{\text{rep}}(S-1)}z_{l+1}, \\
&\vdots \\
0 &= -\frac{L_{\text{via}}}{L_{\text{rep}}}(1 - \epsilon_r)z_0 z_{L_{\text{rep}}} + \frac{z_{L_{\text{rep}}-1}}{L_{\text{rep}}} - \frac{z_{L_{\text{rep}}}}{S-1}.
\end{aligned} \tag{3}$$

There are several features to note about these equations. First, the term, $(L_{\text{via}}/L_{\text{rep}})(1 - \epsilon_r)z_0(1 - z_0)$ and the corresponding terms in the other equations arise from the $\bar{\kappa}(t)$ term in the original quasispecies equations [Eq. (1)] of our model. Second, except for the last equation, the $(l + 1)$st equation has a mutational contribution from z_{l+1} which scales as $1/L_{\text{rep}}$. This means that the contribution to z_l due to backmutation from z_{l+1} becomes negligible for large sequence lengths. This makes sense, since for finite l, the ratio $C_{l'}/C_l \to \infty$ as $L_{\text{rep}} \to \infty$ for $l' > l$, so the probability of mutating to lower values of l vanishes in the limit of infinite sequence length.

We wish to solve these equations for a fixed value of $\alpha \equiv L_{\text{via}}/L_{\text{rep}}$ in the limit of infinite sequence length L. Let us focus first on the behavior of z_0 in this limit. In the first equilibrium equation, the z_1 term drops out as $L_{\text{rep}} \to \infty$, giving

$$0 = z_0[\alpha(1 - \epsilon_r)(1 - z_0) - \epsilon_r], \tag{4}$$

which has the solutions $z_0 = 0, 1 - \epsilon_r/[\alpha(1 - \epsilon_r)]$. The first solution is inconsistent with the requirement that $z_0 = 1$ when $\epsilon_r = 0$. However, the second solution holds only as long as $z_0 \in [0, 1]$. Clearly, $z_0 \leq 1 \forall \epsilon_r$. The other requirement that $z_0 \geq 0$ gives

$$\epsilon_r \leq \frac{\alpha}{1 + \alpha} = \frac{L_{\text{via}}}{L}. \tag{5}$$

Defining $\epsilon_{r,\text{crit}} = \frac{\alpha}{1+\alpha}$, we see that in the limit of infinite genome length, our system has two "phases." For

$\epsilon_r < \epsilon_{r,\mathrm{crit}}$ the system is in a "nonmutator," or, equivalently, "repairer" phase, in which the fraction of nonmutators is a quantity which depends only on α and ϵ_r. At $\epsilon_r = \epsilon_{r,\mathrm{crit}}$ the system undergoes a "phase" transition, which we term the "repair catastrophe," after which the system is in a mutator ("nonrepairer") phase. In this phase, there is essentially no preference for being a nonmutator, and the fraction of nonmutators becomes inversely proportional to the total number of gene sequences.

A key parameter to study in the phase behavior of our model is the localization length, given by

$$\langle l \rangle = \sum_{l=1}^{L_{\mathrm{rep}}} l z_l. \tag{6}$$

This quantity measures the mean Hamming distance of the population from the nonmutator sequence. To compute $\langle l \rangle$ below the phase transition in the limit of $L_{\mathrm{rep}} \to \infty$, we may note that for finite l our equilibrium equations become

$$\begin{aligned} 0 &= \alpha(1-\epsilon_r)z_0(1-z_0) - \epsilon_r z_0, \\ 0 &= -\alpha(1-\epsilon_r)z_0 z_1 + \epsilon_r z_0 - z_1, \\ &\vdots \\ 0 &= -\alpha(1-\epsilon_r)z_0 z_l + z_{l-1} - z_l, \\ &\vdots. \end{aligned} \tag{7}$$

We have already solved the first equation. The next two equations can be solved together to give, for $l \geq 1$,

$$z_l = \epsilon_r[1 + \alpha(1-\epsilon_r)z_0]^{-l} z_0. \tag{8}$$

It should be noted that, while each z_l converges to the corresponding formula given above as $L_{\mathrm{rep}} \to \infty$, the convergence is not uniform, since the larger the l, the larger L_{rep} must be made to get z_l within some specified cutoff of its $L_{\mathrm{rep}} = \infty$ value.

Define $z_{l,\infty} = \lim_{L_{\mathrm{rep}}\to\infty} z_l$. It may be readily checked that $\sum_{l=0}^{\infty} z_{l,\infty} = 1$, so total population is conserved in this limiting process. The localization length is given by

$$\langle l \rangle = \sum_{l=1}^{\infty} l z_{l,\infty} = \frac{1-\epsilon_{r,\mathrm{crit}}}{\epsilon_{r,\mathrm{crit}}} \frac{\epsilon_r}{\epsilon_{r,\mathrm{crit}} - \epsilon_r}. \tag{9}$$

Note that, as expected, the localization length is finite for $\epsilon_r < \epsilon_{r,\mathrm{crit}}$ but diverges at the phase transition.

It is also useful to solve the equilibrium distribution exactly for the case $\alpha = 0$. This corresponds to $L_{\mathrm{rep}} = L$; that is, the entire genome consists of the repair gene. Note that $\epsilon_{r,\mathrm{crit}} = 0$, so that the system is always in the mutator phase. In this case, it may be shown that the equilibrium solution is given by $z_l = \binom{L_{\mathrm{rep}}}{l}(S-1)^l \epsilon_r z_0$ for $l > 0$, and so the requirement that $\sum_{l=0}^{L_{\mathrm{rep}}} z_l = 1$ gives $z_0 = 1/[1 + \epsilon_r(S^{L_{\mathrm{rep}}} - 1)]$. It is readily shown that for large L_{rep}, the localization length $\langle l \rangle \to (1 - 1/S)L_{\mathrm{rep}}$. This result is equivalent to the case of a uniform distribution, which makes sense since for $\alpha = 0$ there is no preference for being a nonmutator in the limit of large L_{rep}.

The phase behavior which emerges from this model may be understood as follows: For highly efficient repair, the selective advantage for being a nonmutator is sufficiently large to cause the population to equilibrate in a localized cluster about the nonmutator sequence. When repair is inefficient, the accumulation of deleterious mutations in both mutators and nonmutators is comparable, and hence the mutators, which are entropically strongly favored, dominate the population. The selective advantage for being a nonmutator is dictated by α, since for low α there is relatively little leakage off of the fitness peak, while for high α there is a large amount of leakage off of the fitness peak. Thus, for low α, repair has to be highly efficient to give the nonmutators a sufficient selective advantage to be in the nonmutator phase, while for high α, nonmutators have a significant advantage even for relatively inefficient repair.

One of the main features to note regarding the mutator-nonmutator equilibrium is that it is independent of the background error probability ϵ. This feature is interesting because as $\epsilon \to 0$, the difference in viability between the mutators and the nonmutators disappears. Thus, one might naively expect $\epsilon_{r,\mathrm{crit}}$ to be a function of ϵ, but in the limit of small ϵ (so that only point mutations are important), this is not the case.

Figure 1 shows a plot of z_0 versus ϵ_r for $\alpha = 1/9$, 1, and 9. We used $S = 2$ and took a value of $L_{\mathrm{rep}} = 1000$ in order to sufficiently converge the calculations. The equilibrium equations were solved by using fixed-point iteration at every ϵ_r. Note that the phase transition does indeed occur at the predicted values of $\epsilon_{r,\mathrm{crit}}$. The analytical $L_{\mathrm{rep}} = \infty$ curves lie essentially on top of our numerical results and were therefore not plotted here.

Figure 2 shows the corresponding plots of $\langle l \rangle$ versus ϵ_r. Note that the localization lengths settle at the value of

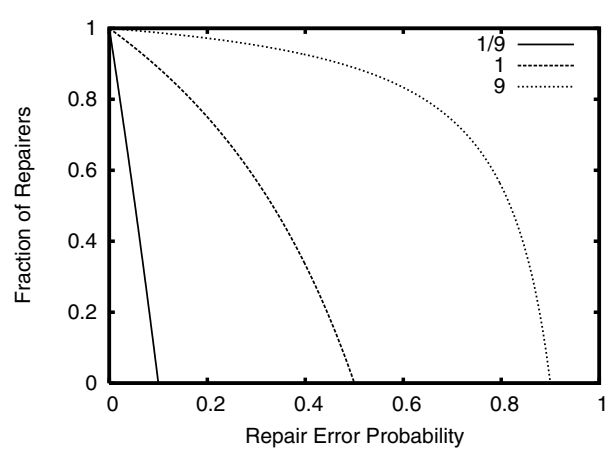

FIG. 1. Plots of z_0 versus ϵ_r for $\alpha = 1/9$, 1, and 9.

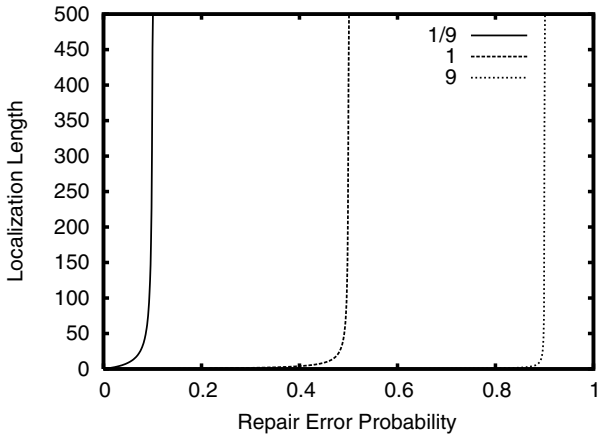

FIG. 2. Plots of $\langle l \rangle$ versus ϵ_r for $\alpha = 1/9$, 1, and 9.

$L_{\rm rep}/2$ at the transition, which is the expected finite $L_{\rm rep}$ behavior for the mutator phase.

Finally, we may use our model to estimate the equilibrium fraction of mismatch repair deficient strains in *E. coli*. The *E. coli* genome has 4×10^6 base pairs, comprising 4403 genes [16]. Based on calculations for *Saccharomyces cerivisiae*, or Baker's yeast, we esimate that between 18%–30% of these genes are "viability" genes, i.e., required for *E. coli* survival [17,18] (unfortunately, similarly detailed data are not currently available for *E. coli*, so we had to make an estimate based on available information). Thus, we assume approximately 1000 viability genes, which we gather into the viability peak of our model. The mismatch repair system is controlled by the MutH, MutL, MutS, and UvrD (or MutU) proteins, giving four repair genes. If we simply use the average gene length and assume that the same fraction of base pairs must be conserved in both the viability genes and in the mismatch repair genes, then in our model we obtain $\alpha \approx 1000/4 = 250$. Since mismatch repair has a failure probability of 10^{-4}–10^{-1} per mismatched base pair, we estimate an equilibrium fraction of mutators in the range of 4×10^{-7}–4×10^{-4}. For *E. coli*, the observed equilibrium fraction of mutators is on the order of 10^{-5}–10^{-3} [8]. While encouraging, our result is nevertheless based on a number of simplifying assumptions. The strongest evidence in support of our model would be the experimental observation of the repair catastrophe itself. While it is not clear how to selectively control the efficiency of the mismatch repair system, if possible this would allow a direct experimental test of our model.

As a concluding remark, we should note that our prediction of a repair catastrophe in mutator-nonmutator equilibrium suggests that phase transitions may underlie the behavior of a variety of biological systems. A classification of the phase behaviors inherent in various biological networks will greatly increase our understanding of the underlying dynamics governing such systems.

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[1] D. Voet and J. Voet, *Biochemistry* (John Wiley and Sons, Inc., New York, NY, 1995), 2nd ed.
[2] M. Eigen, Naturwissenschaften **58**, 465 (1971).
[3] S. Gallucio, Phys. Rev. E **56**, 4526 (1997).
[4] M. Nilsson and N. Snoad, Phys. Rev. E **65**, 031901 (2002).
[5] M. Nilsson and N. Snoad, Phys. Rev. Lett. **84**, 191 (2000).
[6] S. Crotty, C. Cameron, and R. Andino, Proc. Natl. Acad. Sci. U.S.A. **98**, 6895 (2001).
[7] P. Sniegowski, P. Gerrish, and R. Lenski, Nature (London) **387**, 703 (1997).
[8] J. LeClerc, B. Li, W. Payne, and T. Cebula, Science **274**, 1208 (1996).
[9] F. Taddei, M. Radman, J. Maynard-Smith, B. Toupance, P. Gouyon, and B. Godelle, Nature (London) **387**, 700 (1997).
[10] O. Tenaillon, B. Toupance, H. L. Nagard, F. Taddei, and B. Godelle, Genetics **152**, 485 (1999).
[11] A. Giraud, I. Matic, O. Tenaillon, A. Clara, M. Radman, M. Fons, and F. Taddei, Science **291**, 2606 (2001).
[12] I. Matic, M. Radman, F. Taddei, B. Picard, C. Doit, E. Bingen, E. Denamur, and J. Elion, Science **277**, 1833 (1997).
[13] C. Kamp and S. Bornholdt, Phys. Rev. Lett. **88**, 068104 (2002).
[14] C. Kamp, C. O. Wilke, C. Adami, and S. Bornholdt, Complexity **8**, 28 (2002).
[15] L. Mirny and E. Shakhnovich, Annu. Rev. Biophys. Biomol. Struct. **30**, 361 (2001).
[16] F. R. Blattner *et al.*, Science **277**, 1453 (1997).
[17] G. Giaever *et al.*, Nature (London) **418**, 387 (2002).
[18] E. Winzeler *et al.*, Science **285**, 901 (1999).