Evolution of the genomic mutation rate: implications for asexual populations

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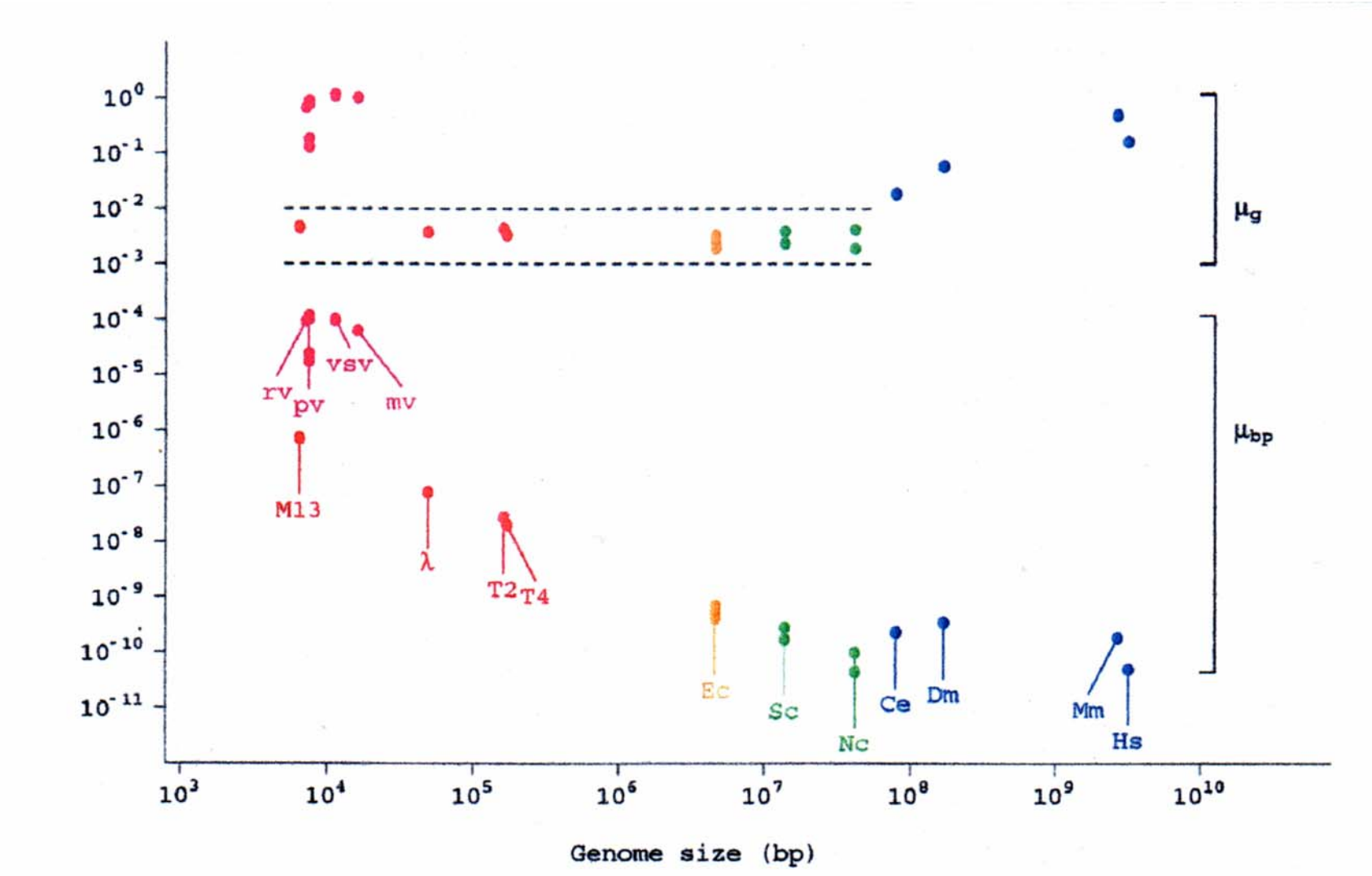
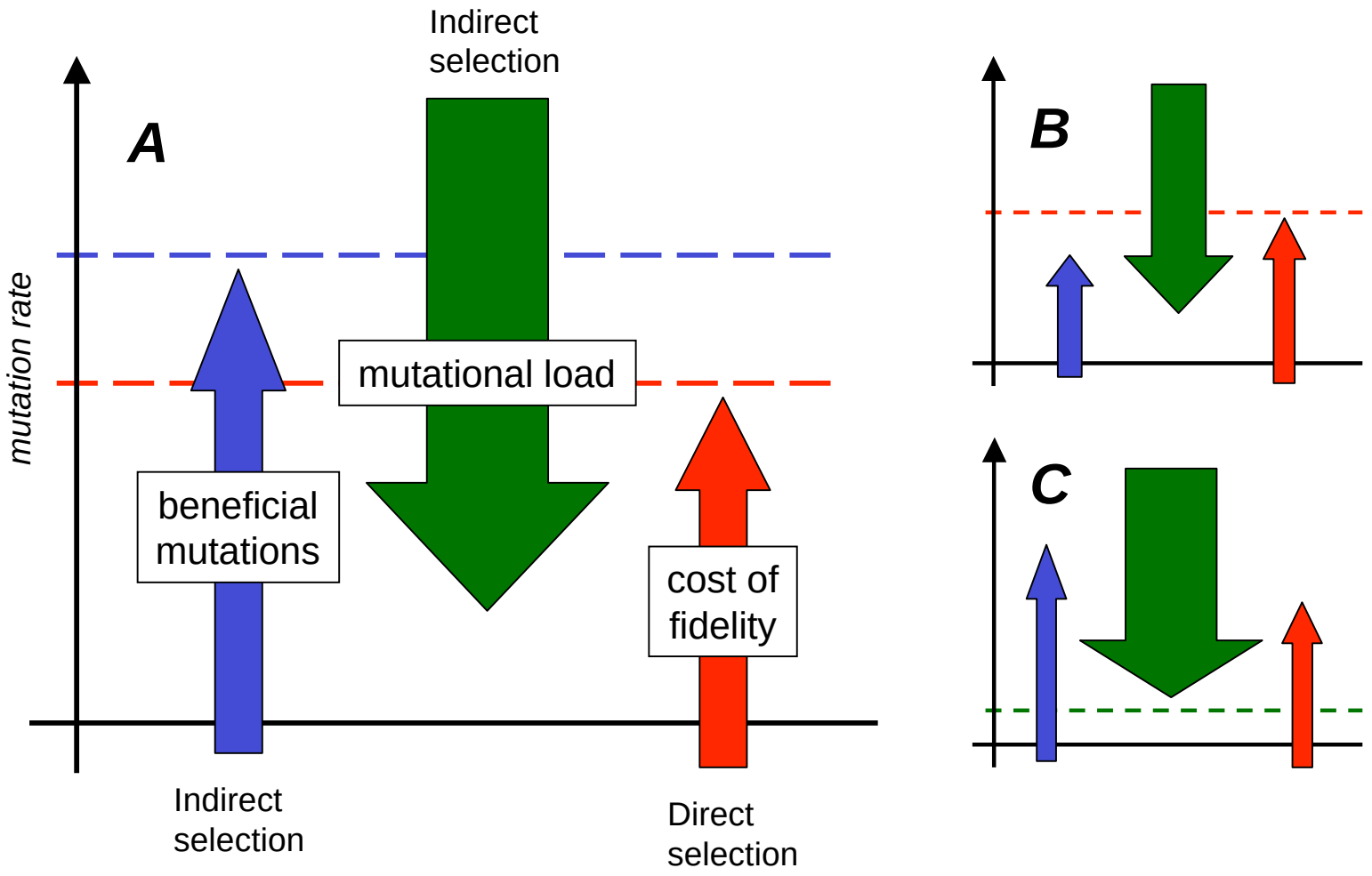
Outline

- Evolution of mutation rate: equilibrium notions
- How does the genomic mutation rate evolve?
- Experimental studies of mutation rate evolution
- The dangers of a high mutation rate for asexuals
- Outline of the “mutation rate catastrophe” hypothesis
- Some theoretical results on the mutation rate ratchet
- Early experimental results
- Implications and future directions

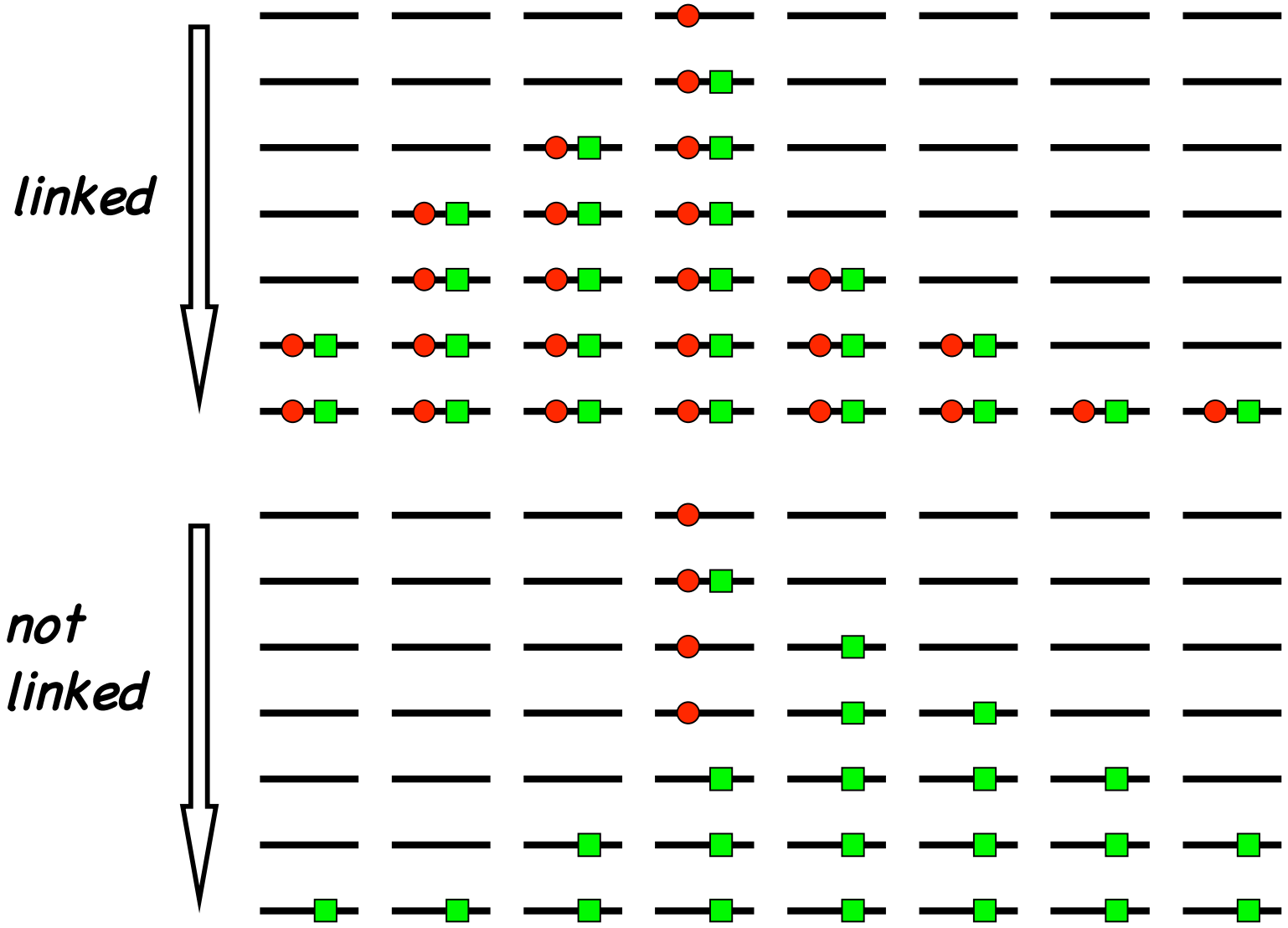
Comparative Data on Mutation Rates



Natural selection and the mutation rate



Linkage and "mutator" hitchhiking



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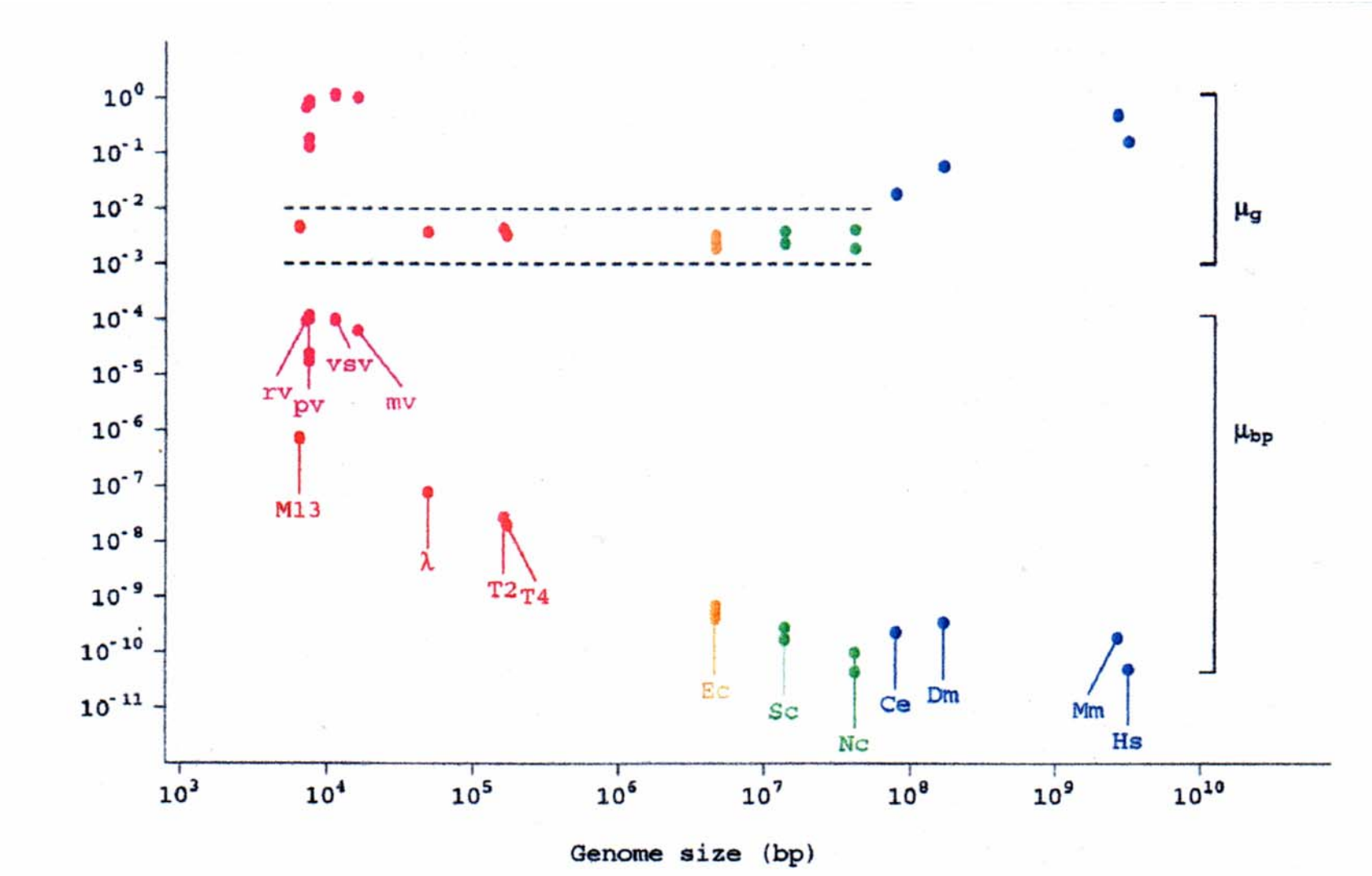
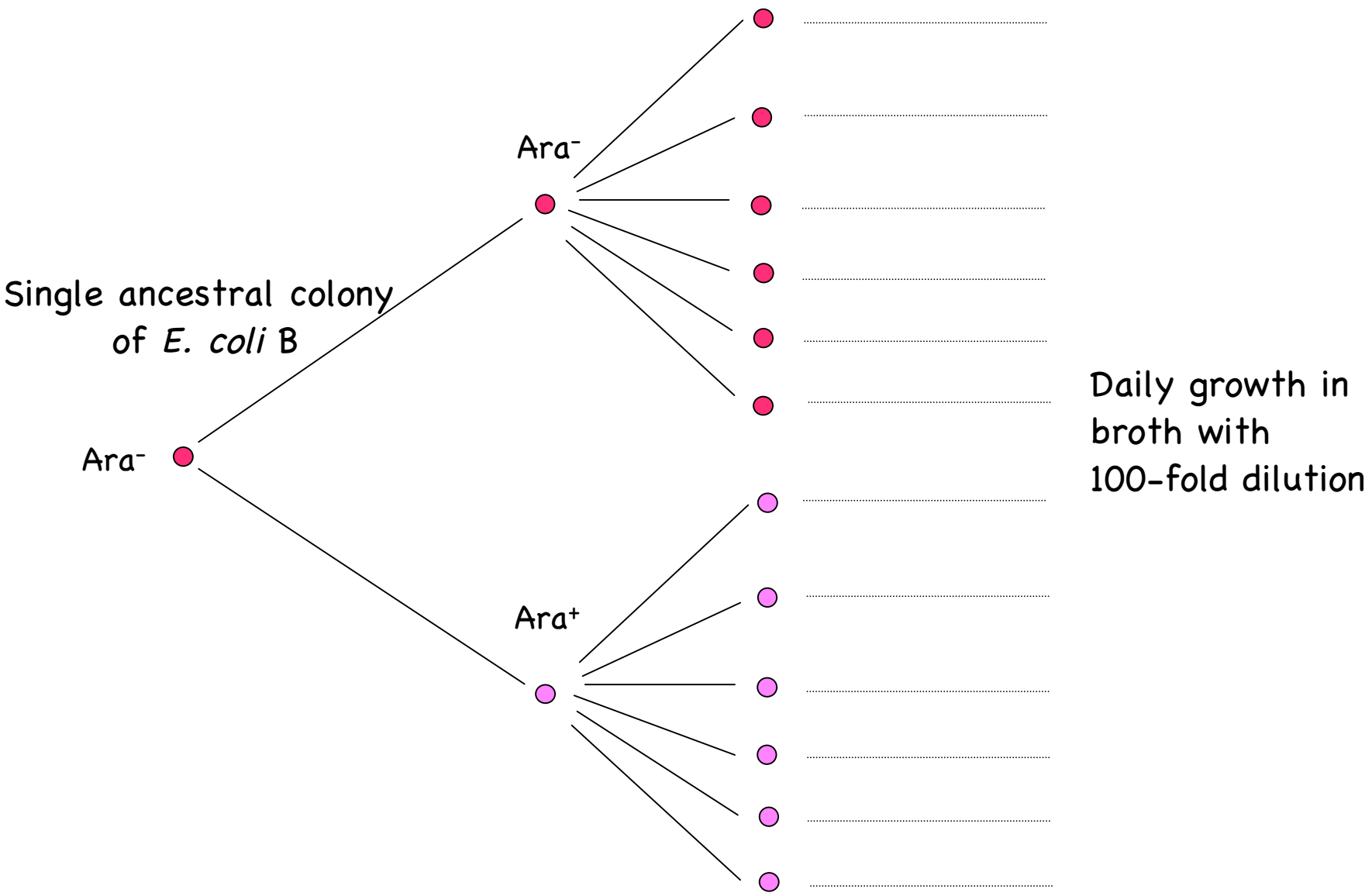


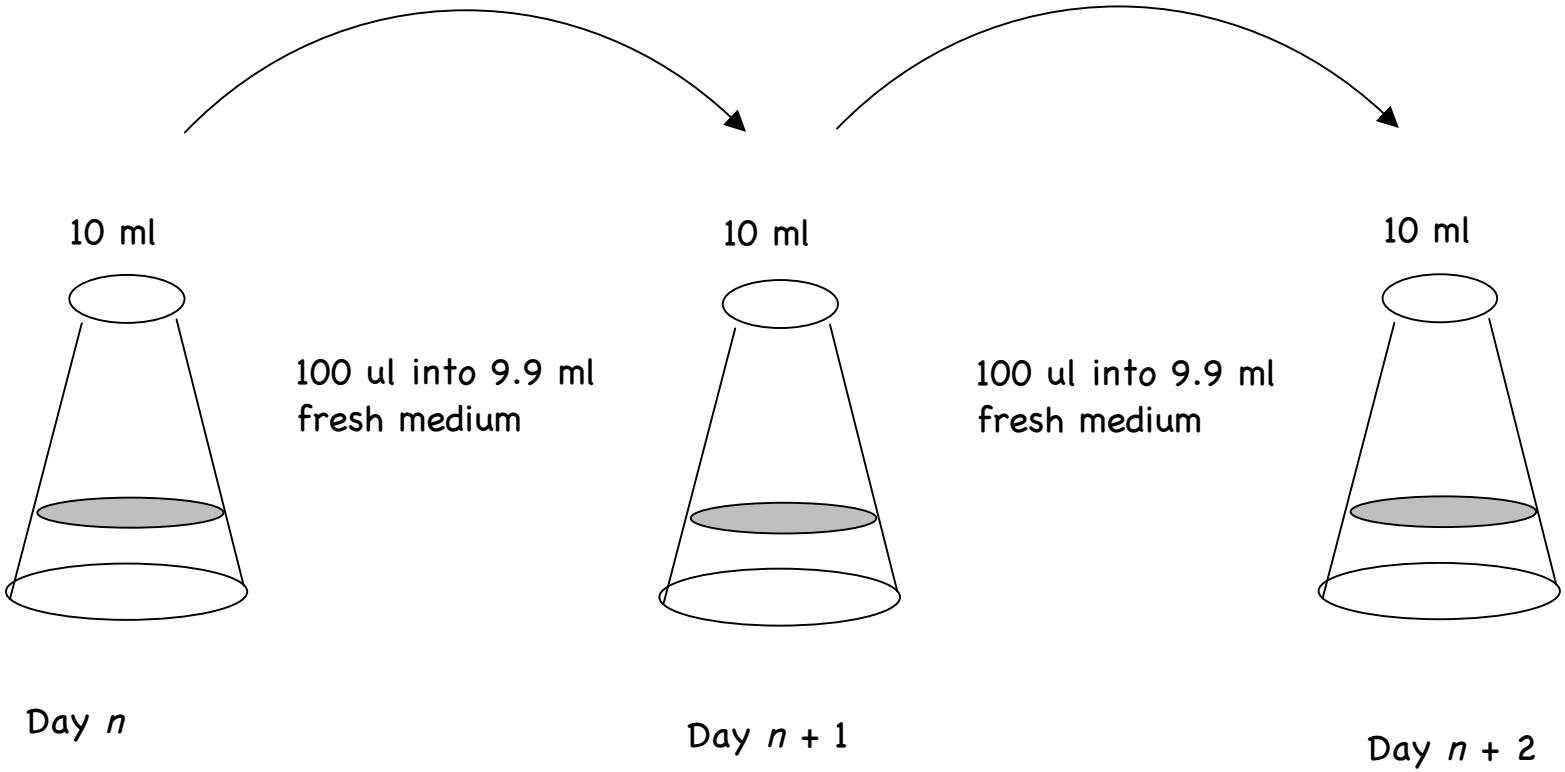
Experimental studies

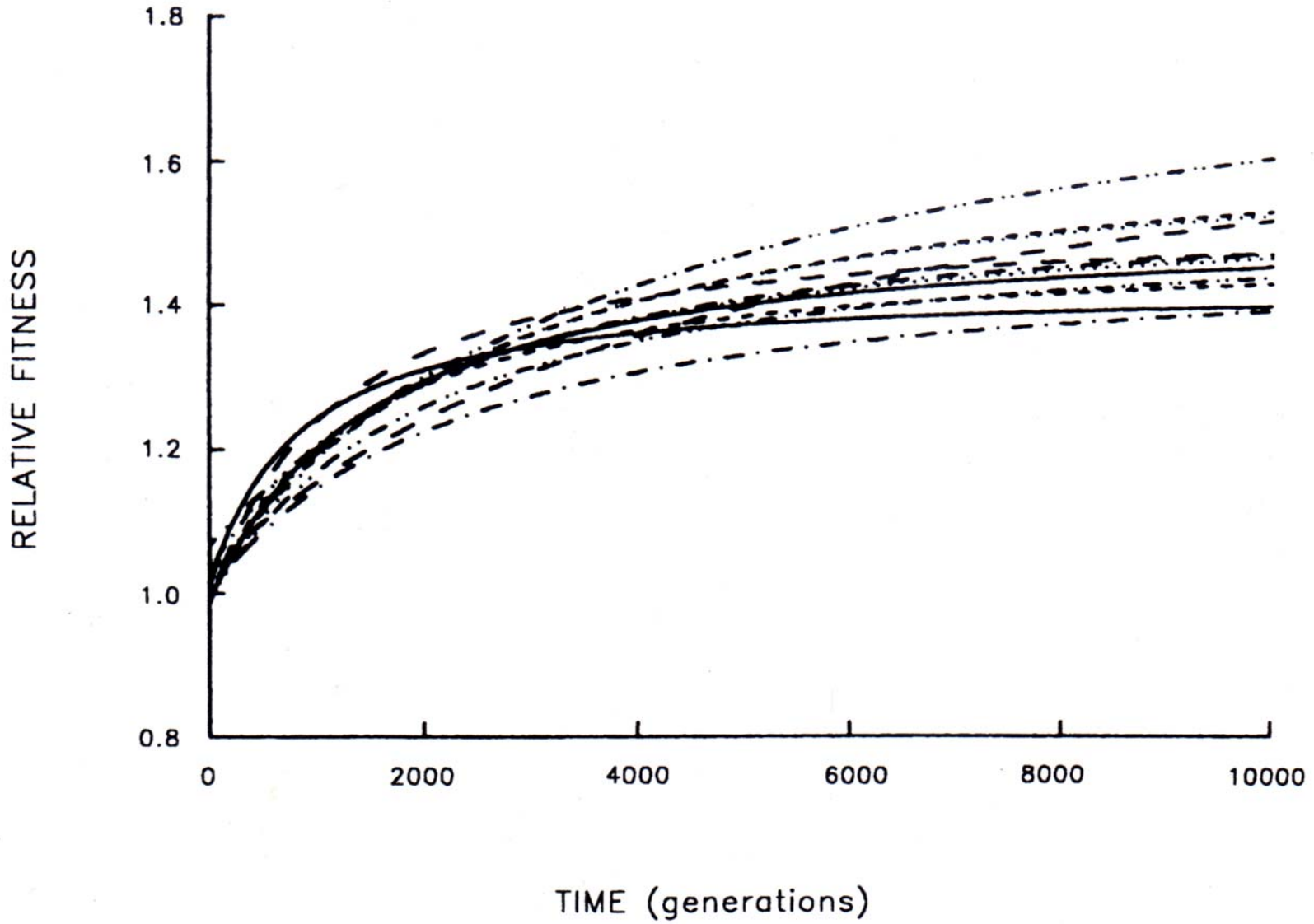
In vitro microbial evolution

Lenski's long term evolution experiment in *E. coli*



Daily transfer regime





Source: Lenski, R.E. and M. Travisano. 1994. *PNAS* 91, 6808-6814.

Generation
0
1000
2000
3000
4000
5000
6000
7000
8000
9000
10000
Arg-5
Arg-4
Arg+3
population
Mutator
Wildtype

Mutator sequence analysis

Population	Mutator mutation	Effect	Additional coding changes at 20,000 generations
Ara^{+}3	insertion of G after position 521	frameshift in 1st third of protein	I->V at a.a. 694, assuming original reading frame
Ara^{-}2	insertion of two a.a. repeat (LA) after a.a. position 68	*see below*	G->E at a.a. 32 M->T at a.a. 135 V->A at a.a. 274
Ara^{-}4	deletion of two a.a. repeat (LA) after a.a. position 68	*see below*	G->D at a.a. 281 A->V at a.a. 606

Note: An LALALA repeat makes up the end of the B a-helix of MutL. Immediately following the repeat is a loop which leads to the C a-helix. This helix-loop-helix structure is thought to form the lid of the ATP binding site for MutL, implying that

Repeat alterations in *mutL*

CTGGCGCTGGCGCTGGCG

Ancestor 68 KKDELALALARHATS

Ara-2 68 KKDELALALALALARHATS

Ara-4 68 KKDELALARHATS

Possible causes of mutator allele substitution

1. Direct selection on mutator allele

   -Predicts advantage of mutator allele over wild type on isogenic background.

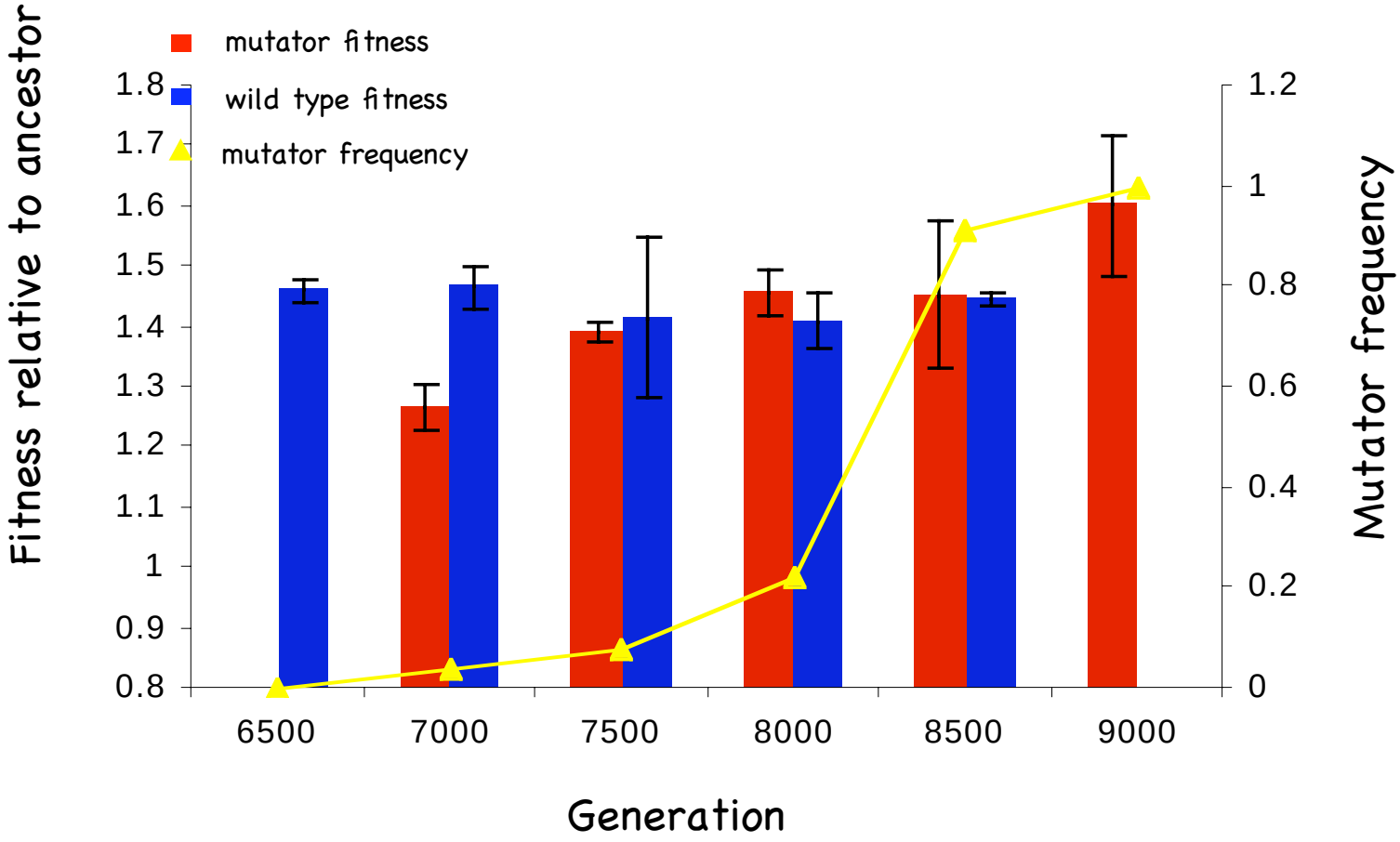
2. Genetic drift

   -Predicts very slow, unsteady rise of allele.

3. Indirect selection (hitchhiking)

   -Predicts fitness gains greater than direct advantage (if any) of allele while the allele is substituting in population.

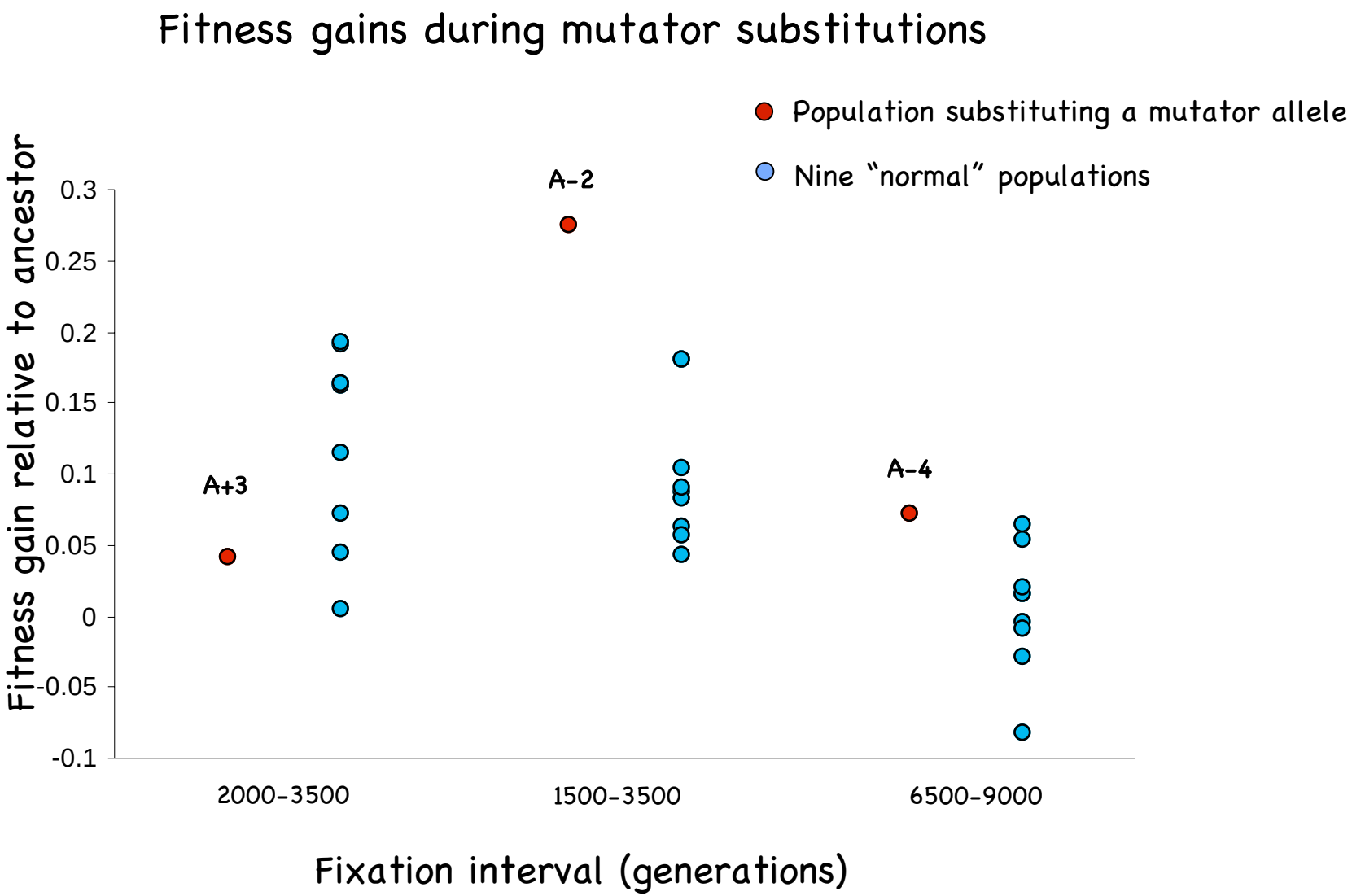
Ara-4 mutator substitution: Average fitnesses



Shaver et al. 2002, *Genetics* 162, 557-566.

$64,000 QUESTION

Is there any adaptive advantage to evolving a high mutation rate?


Fitness gains during mutator substitutions
Population substituting a mutator allele
Nine "normal" populations
Fitness gain relative to ancestor
0.3
0.25
0.2
0.15
0.1
0.05
0
-0.05
-0.1
A+3
A-2
A-4
2000-3500
1500-3500
6500-9000
Fixation interval (generations)

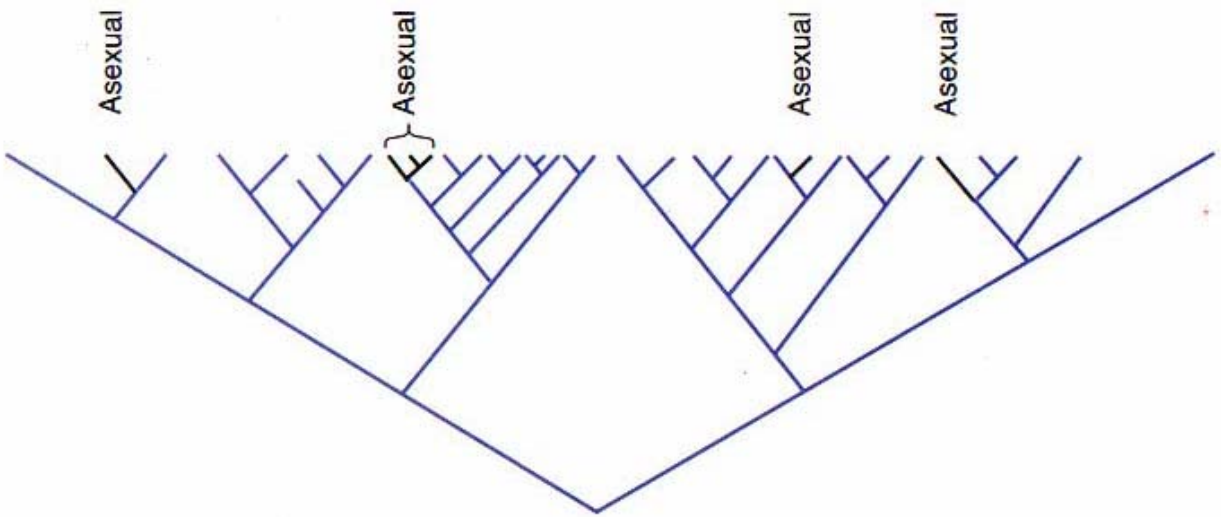
$65,000 QUESTION

What is the ultimate implication of indirect selection on the mutation rate for an asexual population?

(Hint: The answer might be something sinister...)

Asexuals are ephemeral

Figure 11.2 The taxonomic distribution of asexual reproduction is spindly; it is found in odd, isolated taxa.



Source: Ridley, M.2004. *Evolution,* 3rd ed. Blackwell.

Four ways in which an elevated genomic mutation rate can threaten population viability in asexuals

1. Muller's ratchet is accelerated (stochastic).

2. Mutational meltdowns are accelerated (stochastic).

3. The lethal mutation rate is exceeded (deterministic).

4. The error threshold for fitness and/or mutation rate is exceeded (deterministic).

Muller's Ratchet: A cost of asexuality

"...an asexual population incorporates a kind of ratchet mechanism, such that it can never get to contain, in any of its lines, a load of mutations smaller than that existing in its at present least-loaded lines."

Muller 1964

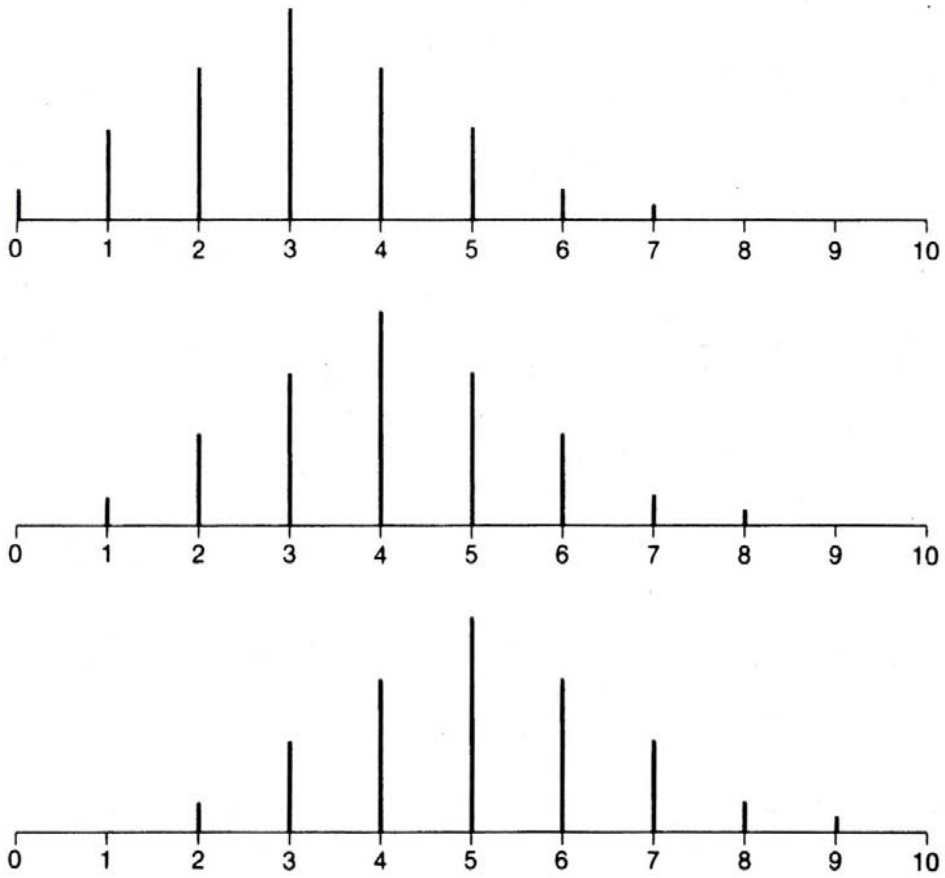


Figure source: Maynard Smith, J. *Evolutionary Genetics*, 2nd ed. Oxford U. Press

Note: Muller's ratchet and mutational meltdowns can be slowed and even stopped by large population size and/or beneficial mutations.

The idea of "lethal mutagenesis" in asexuals (interesting recent papers by Jim Bull, Claus Wilke et al.)

Assume:

- -mutations per genome are Poisson-distributed
- -all mutations deleterious or neutral
- -deleterious mutations arise at genomic rate U_d
- -population infinite

Then mean fitness at equilibrium is the Poisson fraction of mutation-free genotypes:

$$\overline{w} = e^{-U_d}$$

Let b represent the number of offspring that would reproduce in the absence of mutation.

Then extinction in the largest population is assured if equilibrium absolute fitness is less than one:

$$b\, e^{-U_d} < 1$$

Lethal mutagenesis, contd.

Consider a simple model for a bacterial population that in the absence of mutation would be growing exponentially by binary fission (and ignoring absolute generation time). For this situation, $b = 2$ and extinction is assured if

$U_d > 0.69$.

Under more realistic circumstances, a lower genomic rate can cause extinction.

Notes:

--The process is deterministic and does not depend on population size.

--Approach to equilibrium fitness may take many generations and depends on distribution of effects of deleterious mutations. (Equilibrium occurs in 1 generation if all mutations are lethal...) This point about the approach to equilibrium fitness will become important later in a different context.

--Beneficial mutations are not considered in the model but can slow or even prevent extinction.

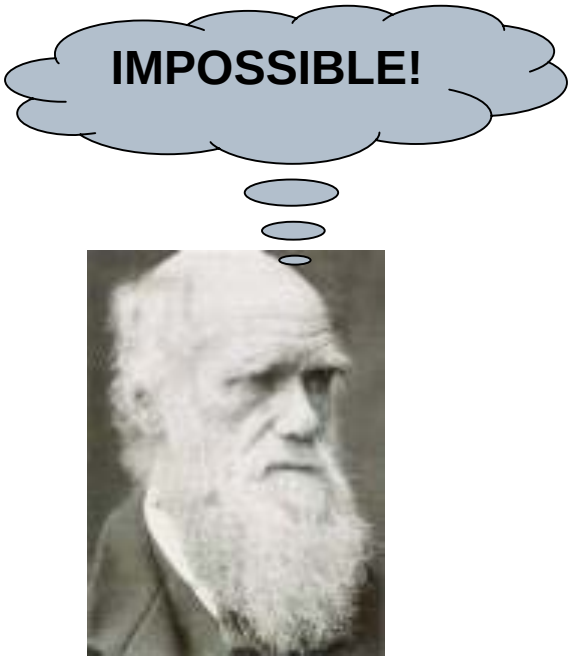
The idea of the "error threshold"

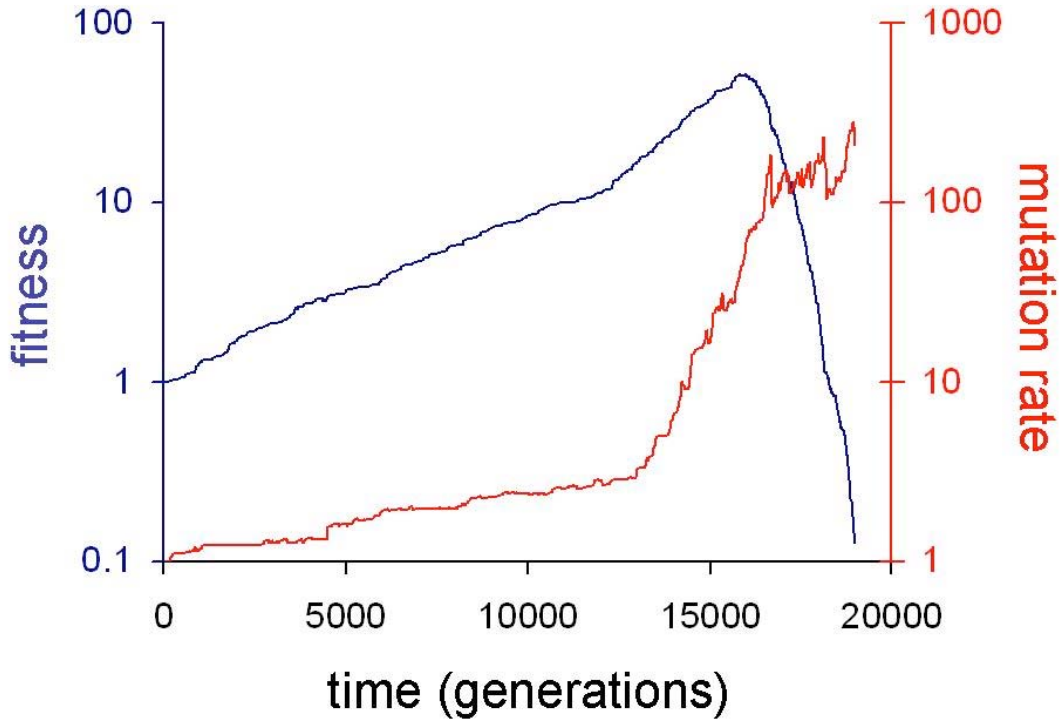
This concept comes from the quasispecies theory of Eigen and Schuster, which considers the evolution of polynucleotide sequences of infinite length in an infinite population under mutation and selection.

For a simple fitness landscape with a single peak, adaptation of the population (by which is meant localization around the peak in sequence space) is impossible if $u > 1/L$, where u is per site mutation rate and L is sequence length. Beyond this threshold the population undergoes a sharp transition to a state in which all sequences become equally likely.

Notes: The significance of the error threshold for extinction is controversial. Its original formulation is for an infinite population, so extinction cannot occur, strictly speaking. The concept has been liberally invoked in the viral mutagenesis literature, but it may be that the theory of lethal mutagenesis is more relevant there. Nonetheless, the error threshold may be an important part of extinction under the mutation rate ratchet process (to be described next), because it is insensitive to the presence of beneficial mutations.

A sinister simulation result





The mutation rate ratchet idea: elements

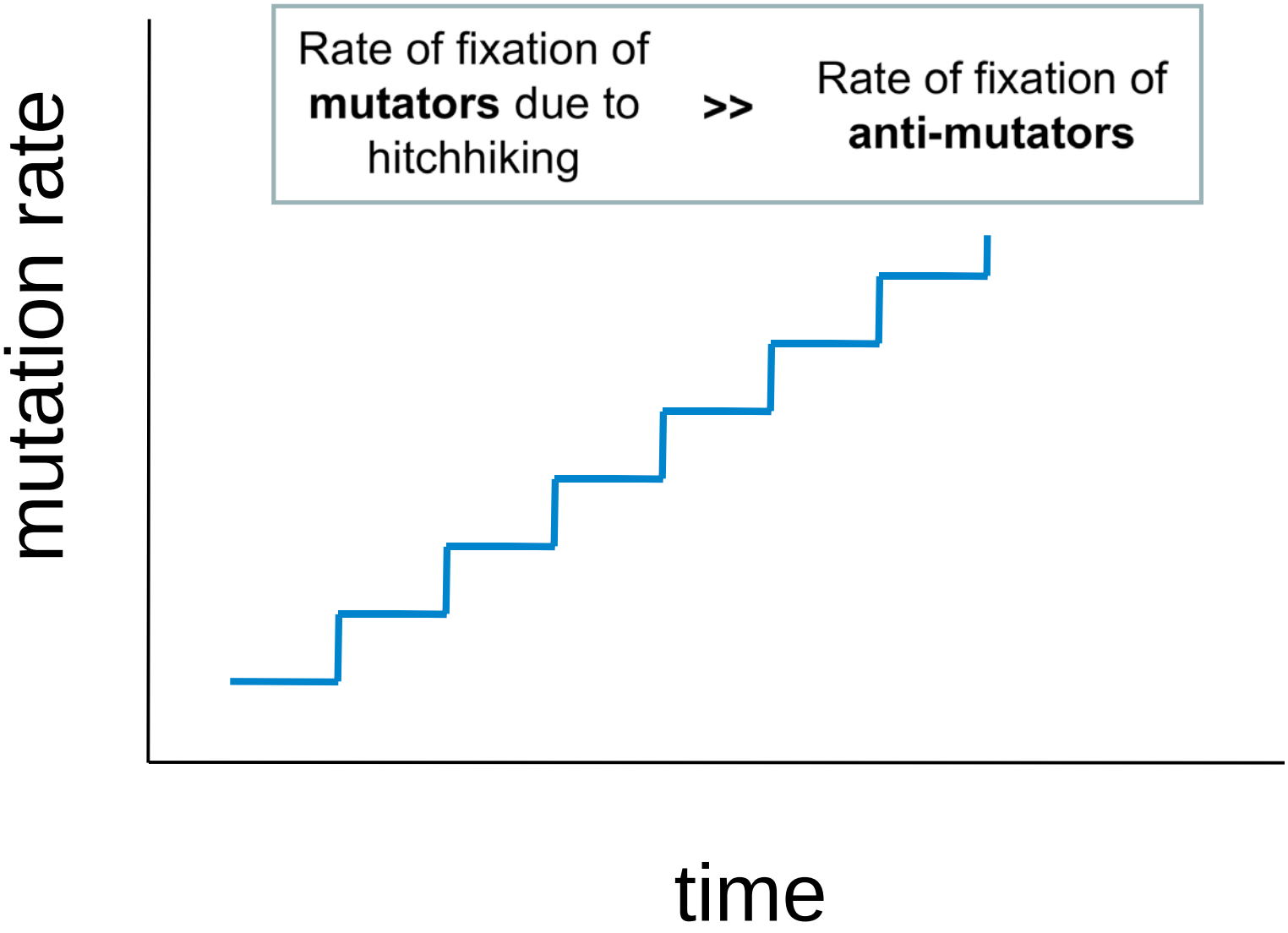
1. The mutation rate in any organism--even viruses--is a polygenic trait. There are many potential mutator loci, and their effects can act cumulatively to produce extremely high mutation rates.

2. Any antimutator allele that arises in a population is most likely to arise on the average loaded background and thus to have no immediate advantage over a mutator allele. Only its future advantage--a decreased accumulation of deleterious mutations over time--is likely to give an antimutator a selective edge. Although the equilibrium cost of a mutator allele in an asexual population is easily calculated and can be substantial, theory indicates that this equilibrium will be approached slowly if there are many mildly deleterious mutations.

3. In the meantime, additional mutator hitchhiking events may occur in association with beneficial mutations, driving the genomic mutation rate even higher.

4. Above a certain genomic mutation rate, the accumulation of deleterious mutations causes the extinction of an asexual population in the relatively short term.

Mutation-rate ratchet



The mutation rate *catastrophe* hypothesis

Our hypothesis is that recurrent mutator hitchhiking (the mutation rate ratchet) can take an asexual population beyond the tolerable mutation rate, causing extinction.

We will show two kinds of results that support this possibility: Individual-based simulations and numerical solution of a PDE model.

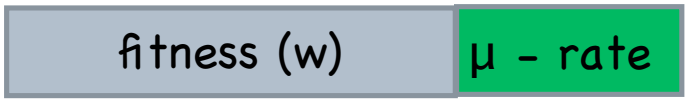
The required linkage

Loci that affect mutation rate: replication, repair, proofreading, etc…

Loci that affect fitness: use your imagination…

Simulations

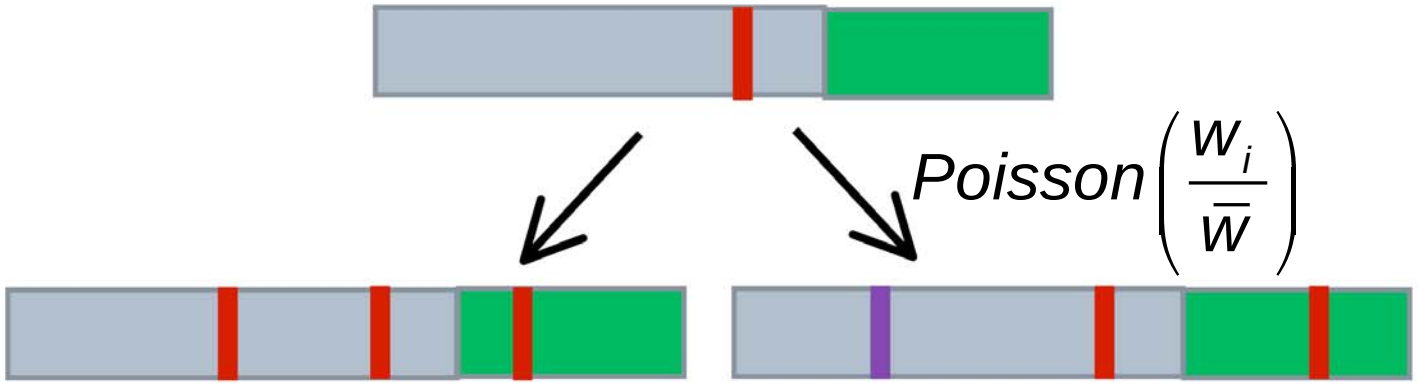
genome



↑ entropy: ↓ w, ↑ μ

↓ entropy: ↑ w, ↓ μ

Replication:



Simulating mutation rate evolution in asexuals

-Simulations keep track of every individual and every replication event in the population.

-Populations are haploid and asexual.

-Offspring can differ from parents in fitness, in mutation rate, or both.

-Offspring number is Poisson distributed for each individual; the mean is set by the fitness of the individual relative to the population mean.

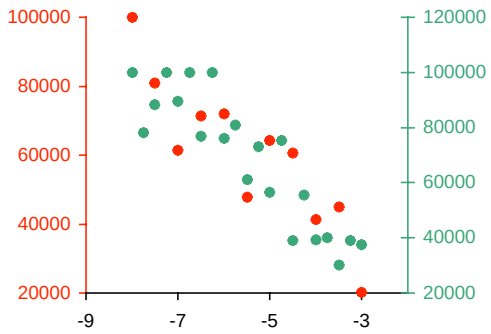
-Offspring acquire X_D new deleterious mutations that are either lethal or decrease fitness by a factor $(1 - M_D)$ and X_B new beneficial mutations that increase fitness by a factor $(1 + M_B)$.

-Offspring acquire X_M new mutator mutations that increase log mutation rate by M_M and X_A new antimutator mutations that decrease log mutation rate by M_A.

-X_D, X_B, X_M, and X_A are all Poisson random variables with means μ_D, μ_B, μ_M, and μ_A that depend on the genomic mutation rate, the relative genomic mutation rate of an individual (which mutates and evolves...), and the fractions of deleterious, beneficial, mutator, and antimutator mutations f_D, f_B, f_M, and f_A, which are set by the user.

-M_D, M_B, M_M, and M_A are continuous random variables with means m_D, m_B, m_M, and m_A; a wide variety of distributions of these mutational effects was explored.

A



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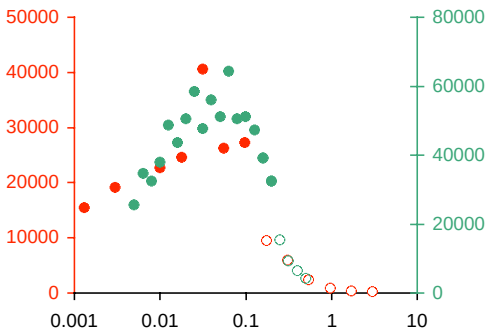

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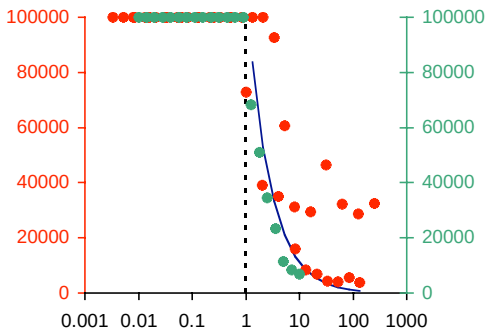

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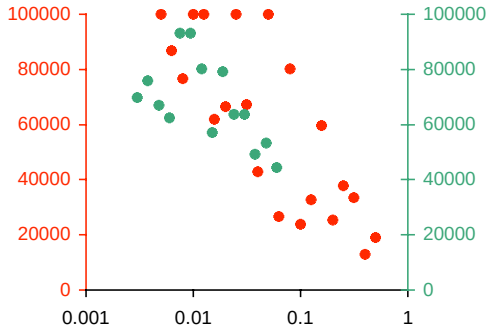

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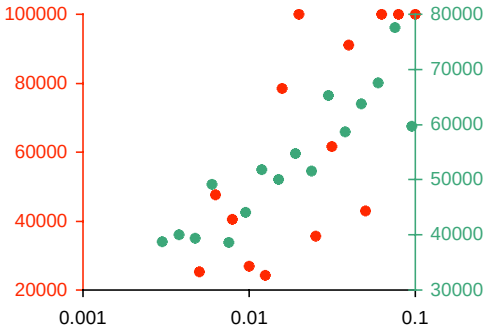

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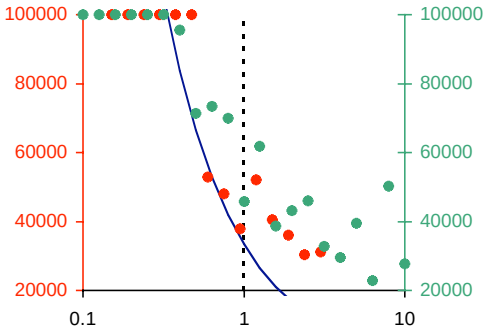

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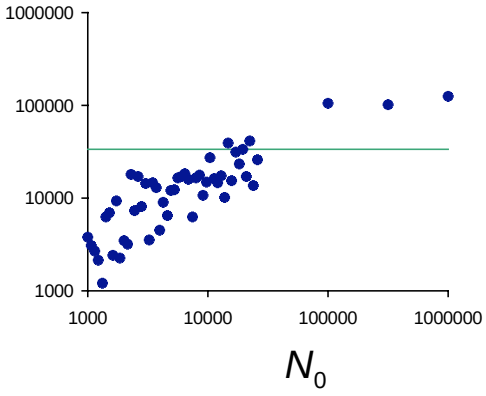


Default parameter values

μ = 0.1
f_B = 0.0003
f_D = 0.1
f_M = 0.001
f_A = 0.0001
$m_B = m_d = m_M = m_A$ = 0.03
N = 10.000

Analytical theory

Model 1

$$\frac{\partial u}{\partial t} = (x - \bar{x})u + e^{y}\mathbf{M}u$$

Model 2

$$\frac{\partial u}{\partial t} = (x - \bar{x})(u + e^{y}\mathbf{M}u)$$

$u = u(x,y,t)$, the probability density of genotypes with fitness x and mutation rate y at time t.

M is the mutation operator and models mutation to and from other genotypes.

$$e^{y}\mathbf{M}u = f_B e^{y}\left(\int_0^{\infty} u(x-\Delta x, y, t)\, g_B(\Delta x)\, d\Delta x - u\right) + f_D e^{y}\left(\int_0^{\infty} u(x+\Delta x, y, t)\, g_D(\Delta x)\, d\Delta x - u\right) +$$

$$f_M\left(\int_0^{\infty} e^{y-\Delta y} u(x, y-\Delta y, t)\, g_M(\Delta y)\, d\Delta y - e^{y}u\right) + f_{AM}\left(\int_0^{\infty} e^{y+\Delta y} u(x, y+\Delta y, t)\, g_{AM}(\Delta y)\, d\Delta y - e^{y}u\right)$$

f_B = fraction of mutations that increase fitness (beneficial)
f_D = fraction of mutations that decrease fitness (deleterious)
f_M = fraction of mutations that increase mutation rate (mutator)
f_{AM} = fraction of mutations that decrease mutation rate (antimutator)
g_B = distribution of effects for beneficial mutations
g_D = distribution of effects for deleterious mutations
g_M = distribution of effects for mutator mutations
g_{AM} = distribution of effects for antimutator mutations

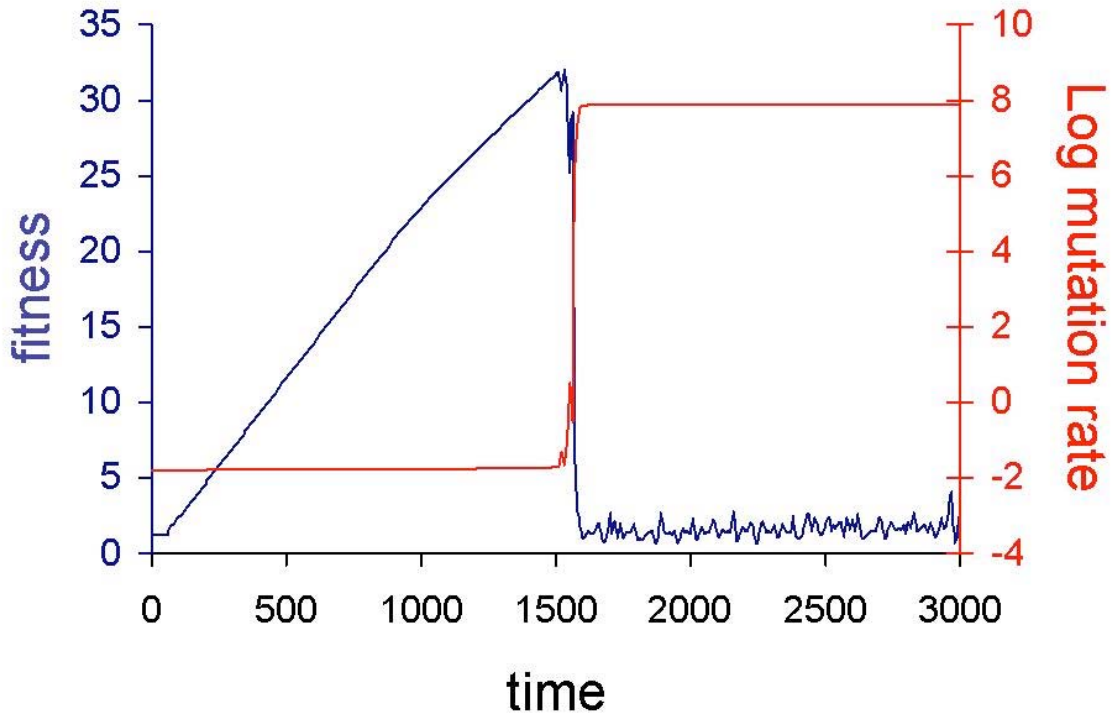
$$\mathbf{M}u \approx D_x \frac{\partial^2 u}{\partial x^2} + d_x \frac{\partial u}{\partial x} + D_y \frac{\partial^2 u}{\partial y^2} + (2D_y + d_y)\frac{\partial u}{\partial y} + (D_y + d_y)\, u$$

$$D_x = \tfrac{1}{2} f_D (m_D^2 + \sigma_D^2) + \tfrac{1}{2} f_B (m_B^2 + \sigma_B^2) \qquad d_x = f_D m_D - f_B m_B$$

$$D_y = \tfrac{1}{2} f_A (m_A^2 + \sigma_A^2) + \tfrac{1}{2} f_M (m_M^2 + \sigma_M^2) \qquad d_y = f_A m_A - f_M m_M$$

Numerical solution of Model 1

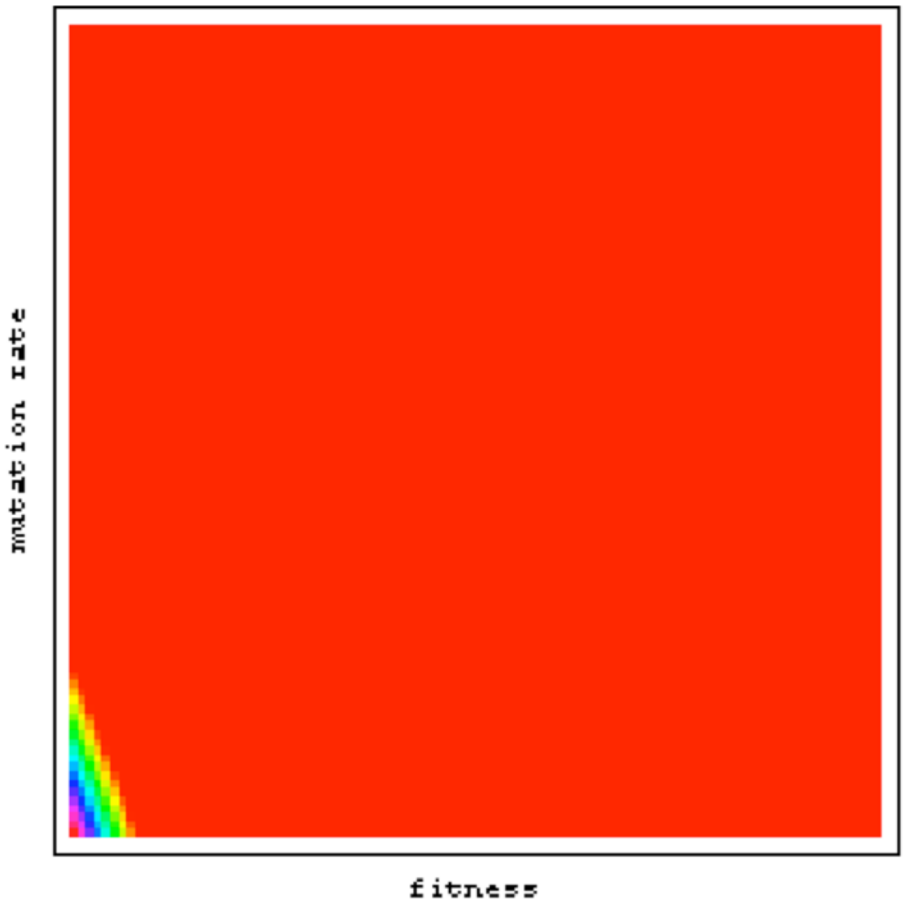
μ = 0.003
f_B = 1e-6
f_D = 0.1
f_M = 0.001
f_A = 0.0001
$m_B = m_D = m_M = m_A$ = 0.003



Numerical solution

- contour plot on a log scale.
- fitness increase "pulls" mutation rate up.
- eventual "phase transition"
- final state: highest mutation rate, lowest fitness.

f_B = 1e-10
f_D = 1e-2
f_M = 1e-4
f_A = 1e-5
$m_B = m_D = m_M = m_A = 0.03$



Conditions for extinction under the mutation rate ratchet (mutation rate catastrophe): Extinction occurs

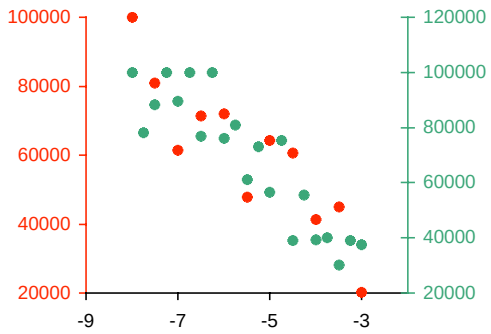
IF: $f_M > f_{AM}$

(mutator mutations more common than antimutator mutations)

IFF: $f_D > f_B > 0$

(deleterious mutations more common than beneficial mutations)

A



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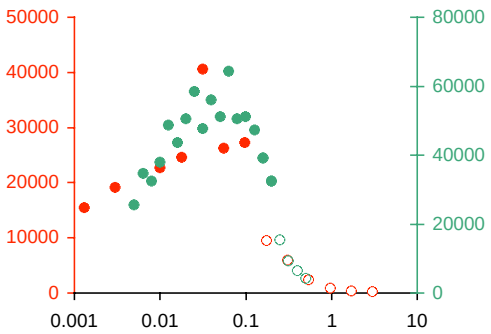

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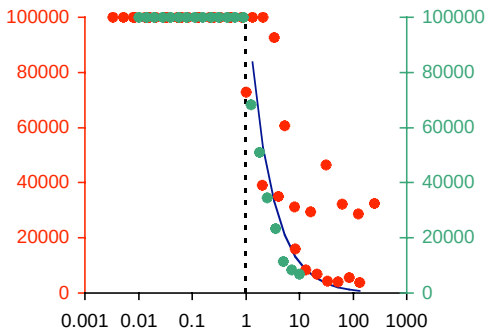

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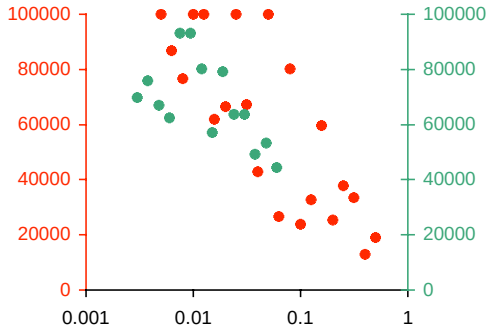

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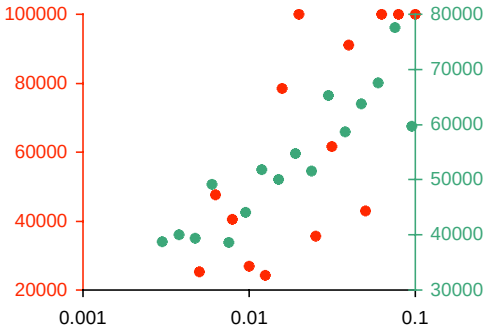

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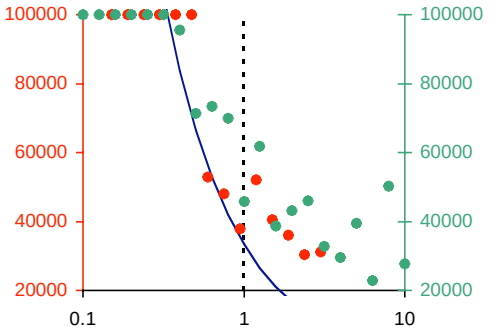

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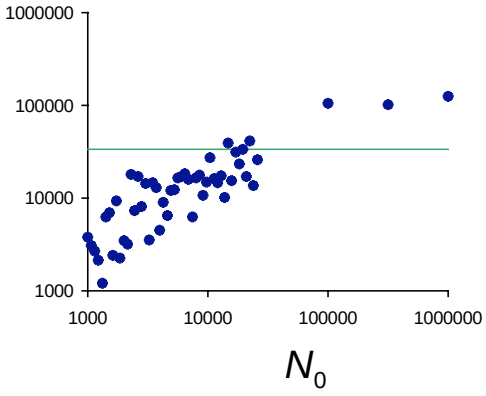

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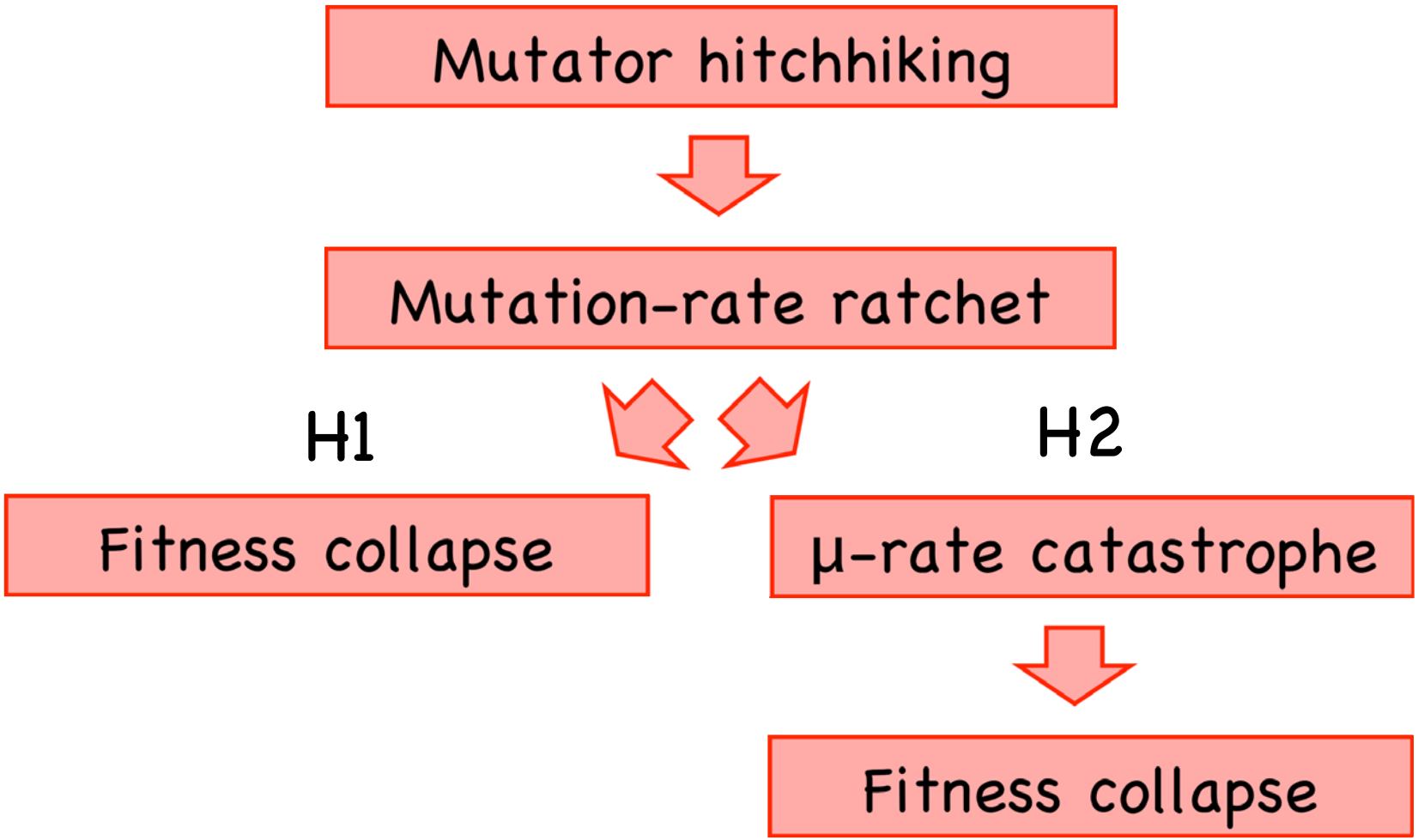
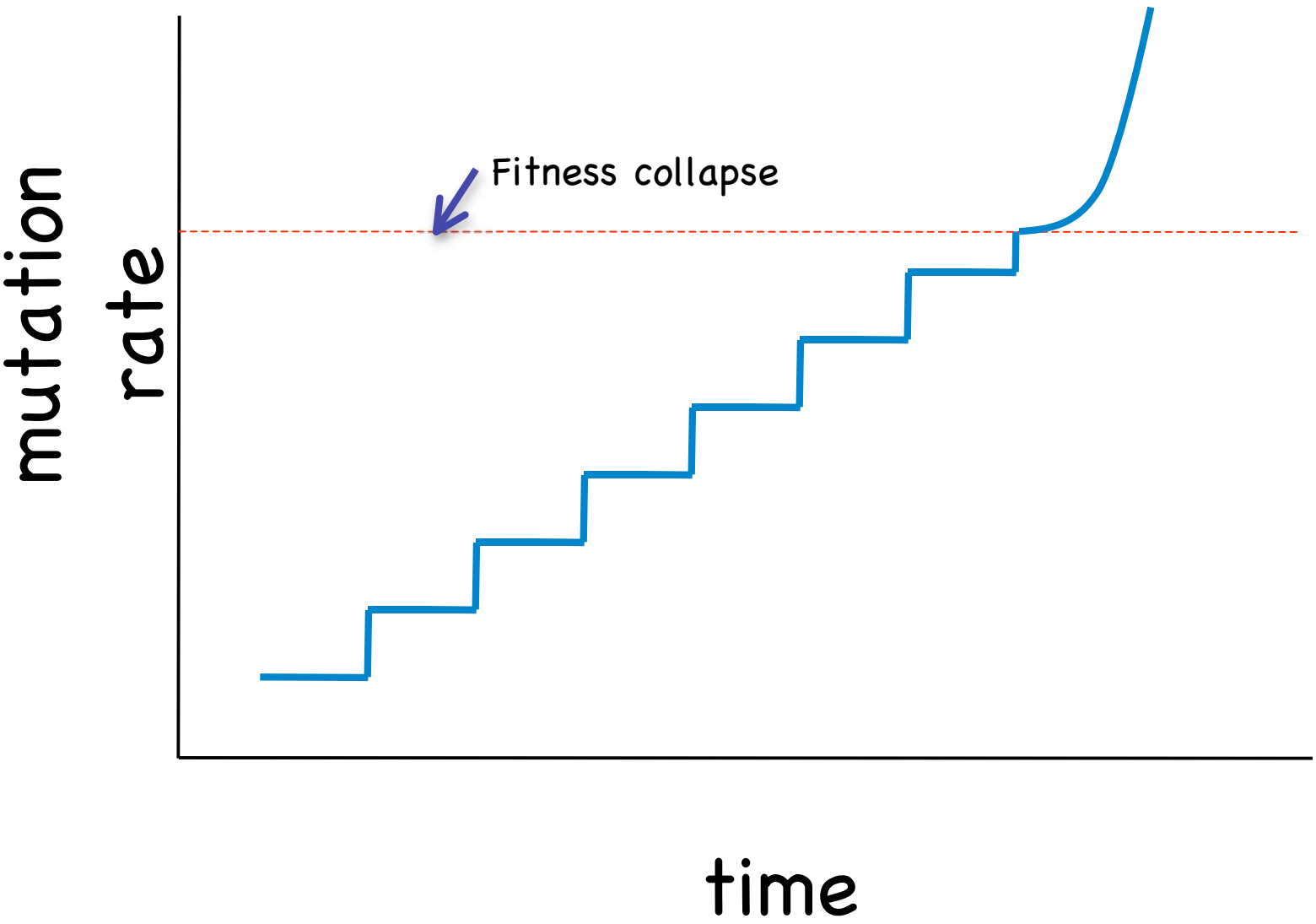
How it might work



How it works: one possibility

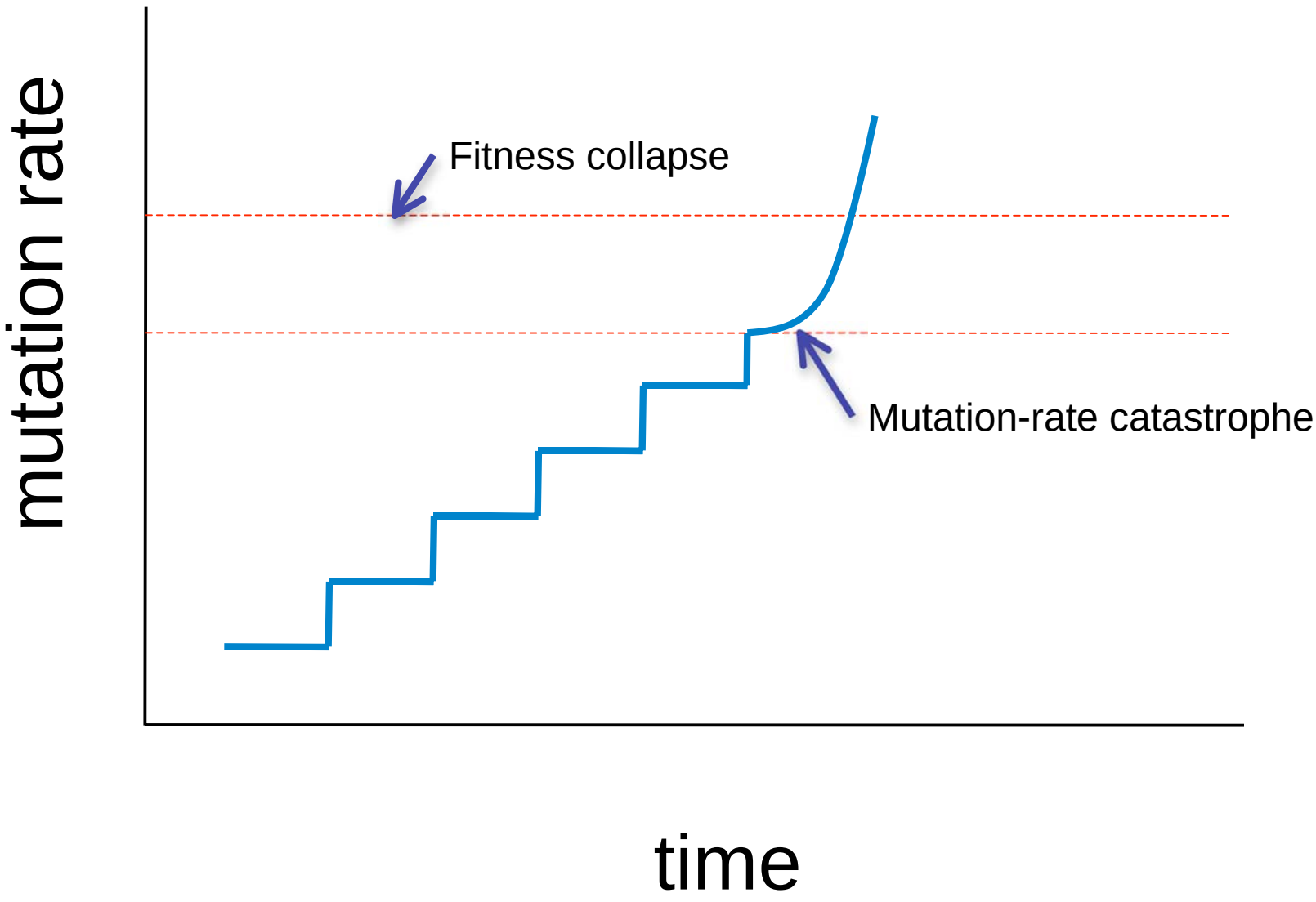
H1: Fitness collapse first


mutation rate
Fitness collapse
time

How it works: the other possibility

H2: Mutation-rate catastrophe

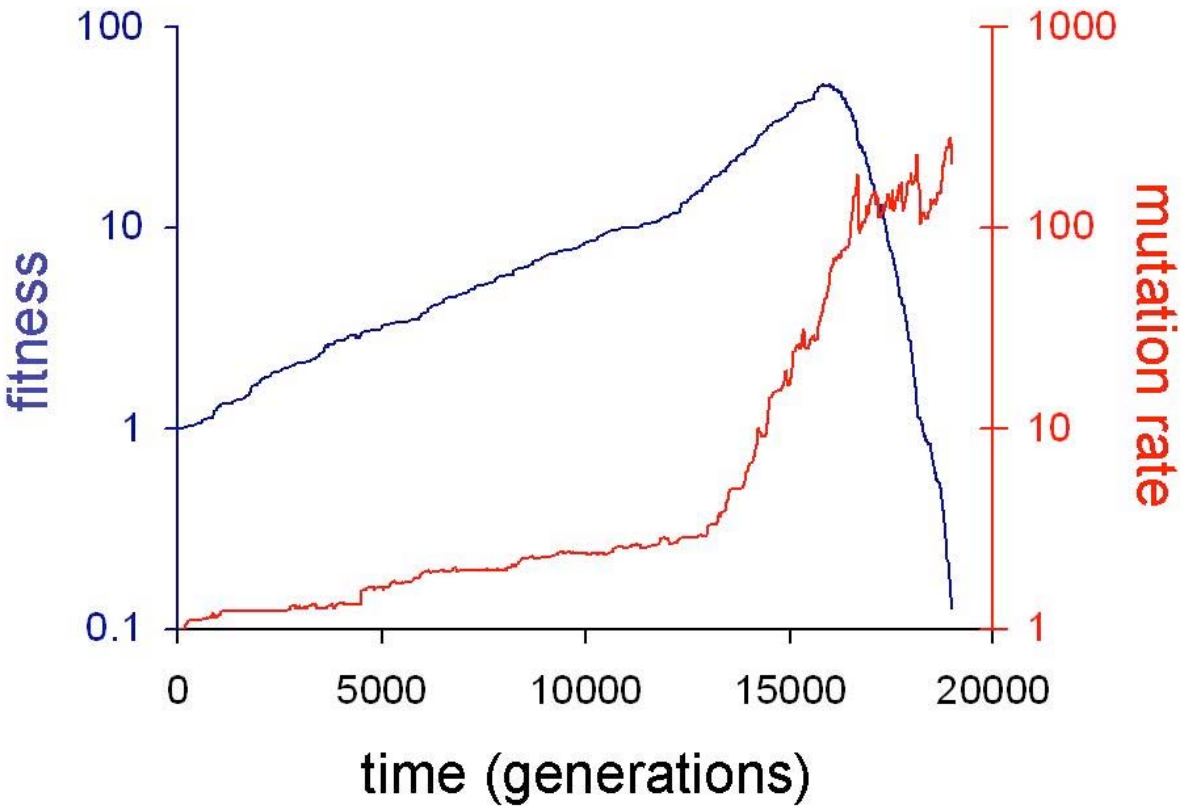
Fitness collapse


mutation rate
Fitness collapse
Mutation-rate catastrophe
time

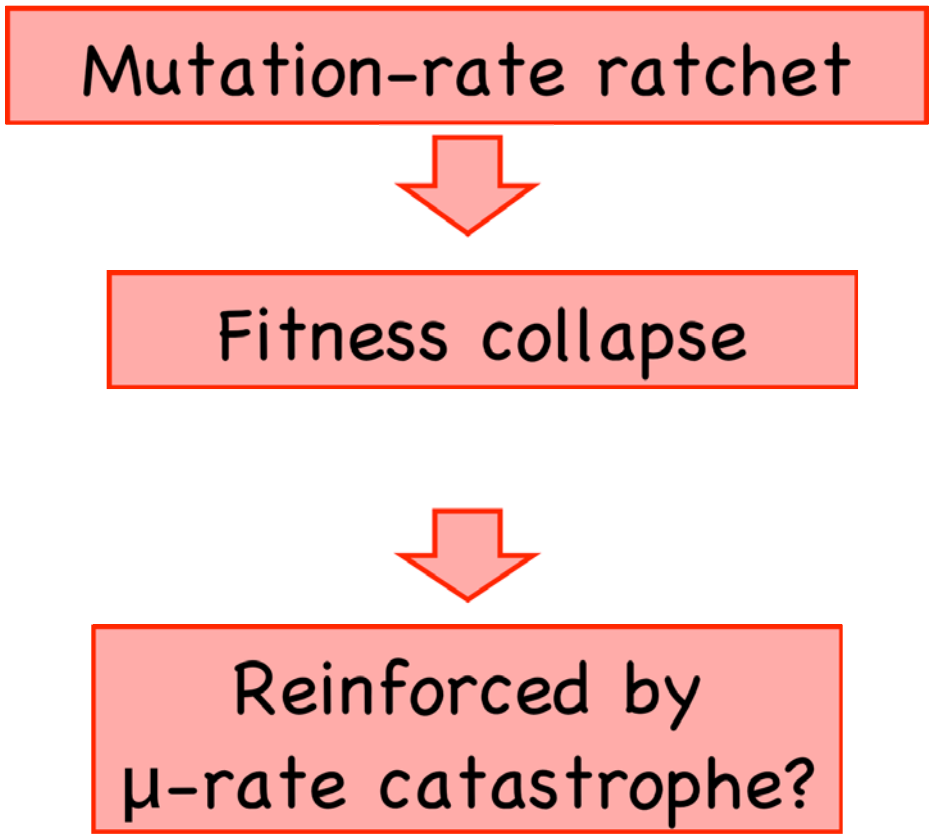
H1 or H2?

What the simulation evidence seems to support.

Fitness peaks first, and mutation rate keeps rising



The emerging picture: H1



Why it works (in simulations, at least)

Why does natural selection, a process that operates through the avoidance of extinction, drive a population extinct?

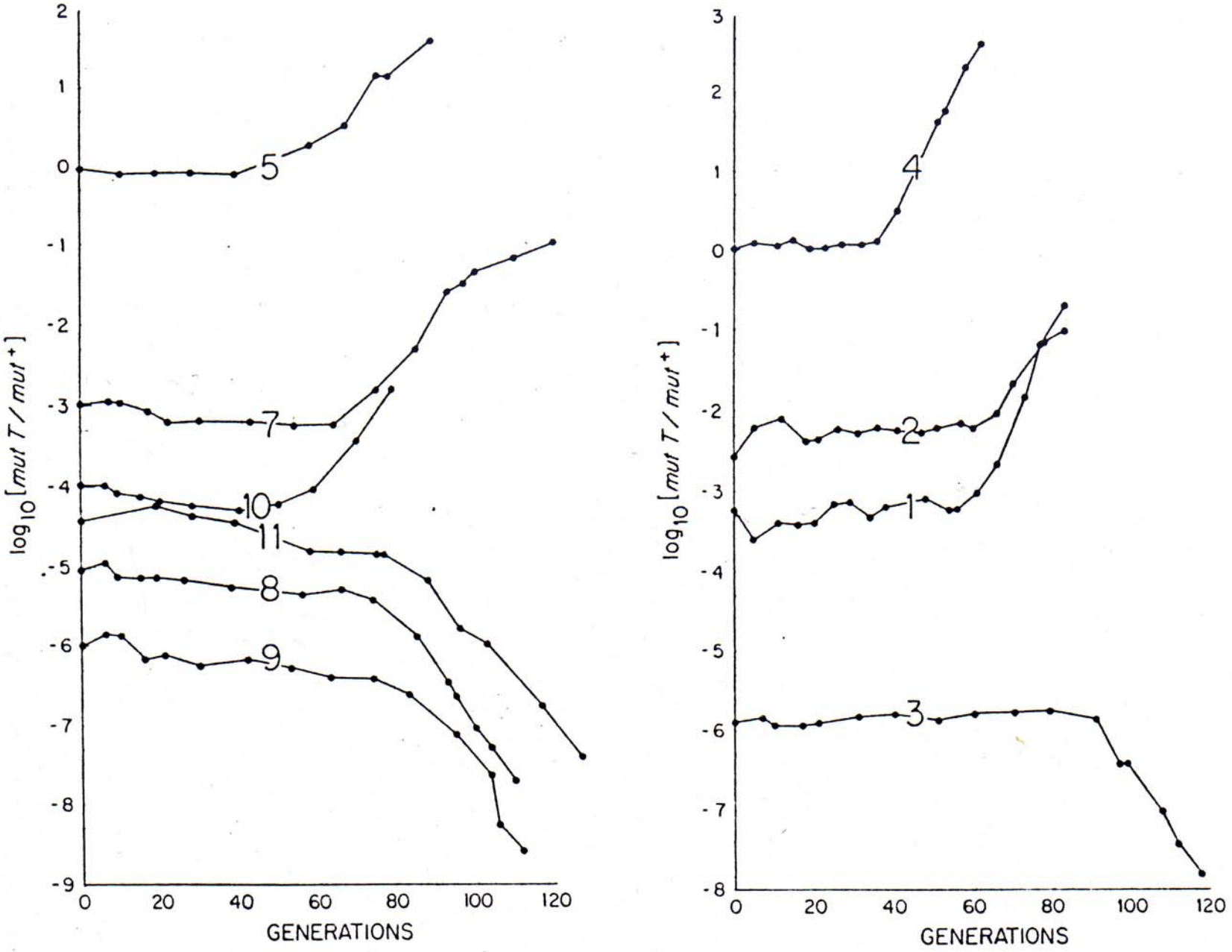
Natural selection is short-sighted

- Mutational load takes time to accumulate.
- Natural selection does not anticipate the realization of excess mutational load.

Courtesy of Warner Bros. Entertainment Inc.

How to test it experimentally?

- Can't wait for whole process: too long.
- However, mutator hitchhiking *is* predictable in short run (to be addressed in a minute).
- A possible approach:
  - Create strains with one and two mutator defects. (Done.)
  - Test for hitchhiking of double mutator over single; this is one click of the mutator ratchet. (Done.)
  - Establish condition under which double-mutator (but not single!) has intolerable mutation rate. See if double-mutator genotype hitchhikes to fixation, then the population goes extinct.



Source: Chao, L. and E.C. Cox. 1983. *Evolution* 37, 125-134

Single and double-mutator strains: mutation rates

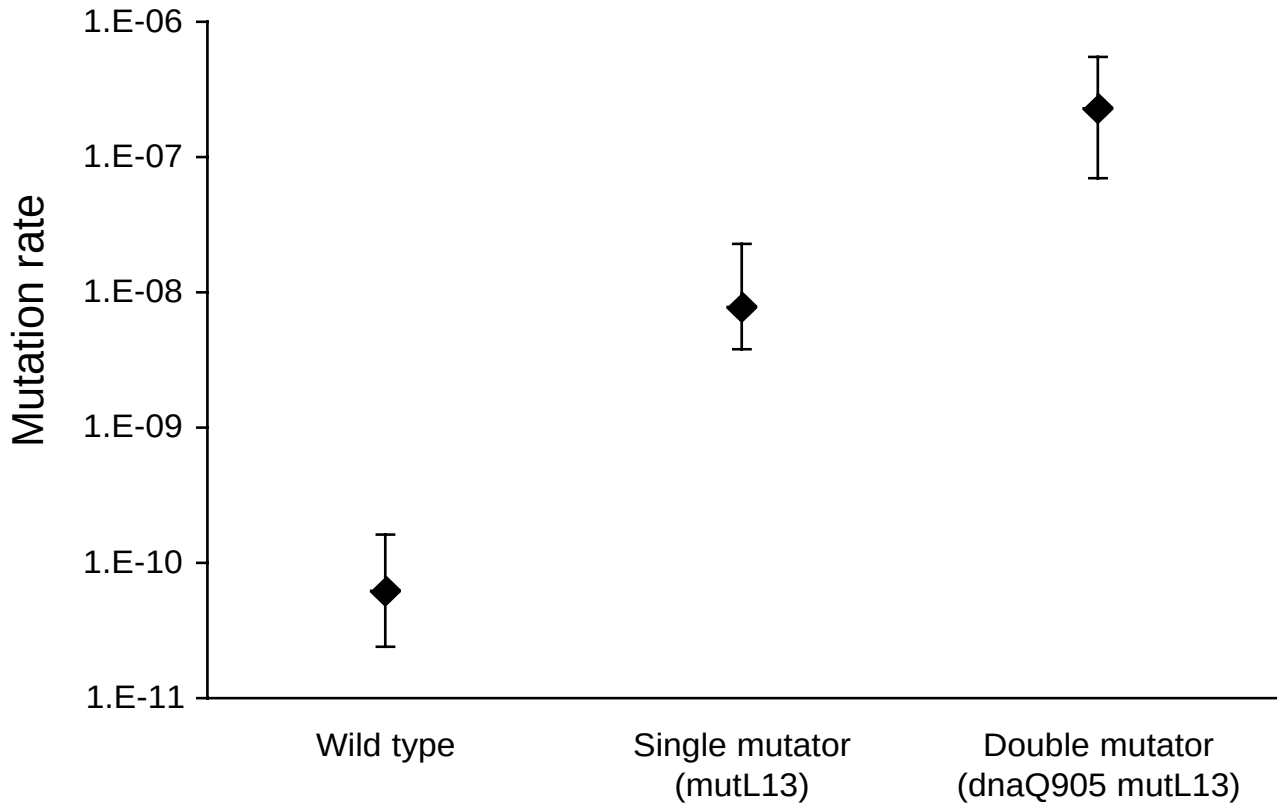
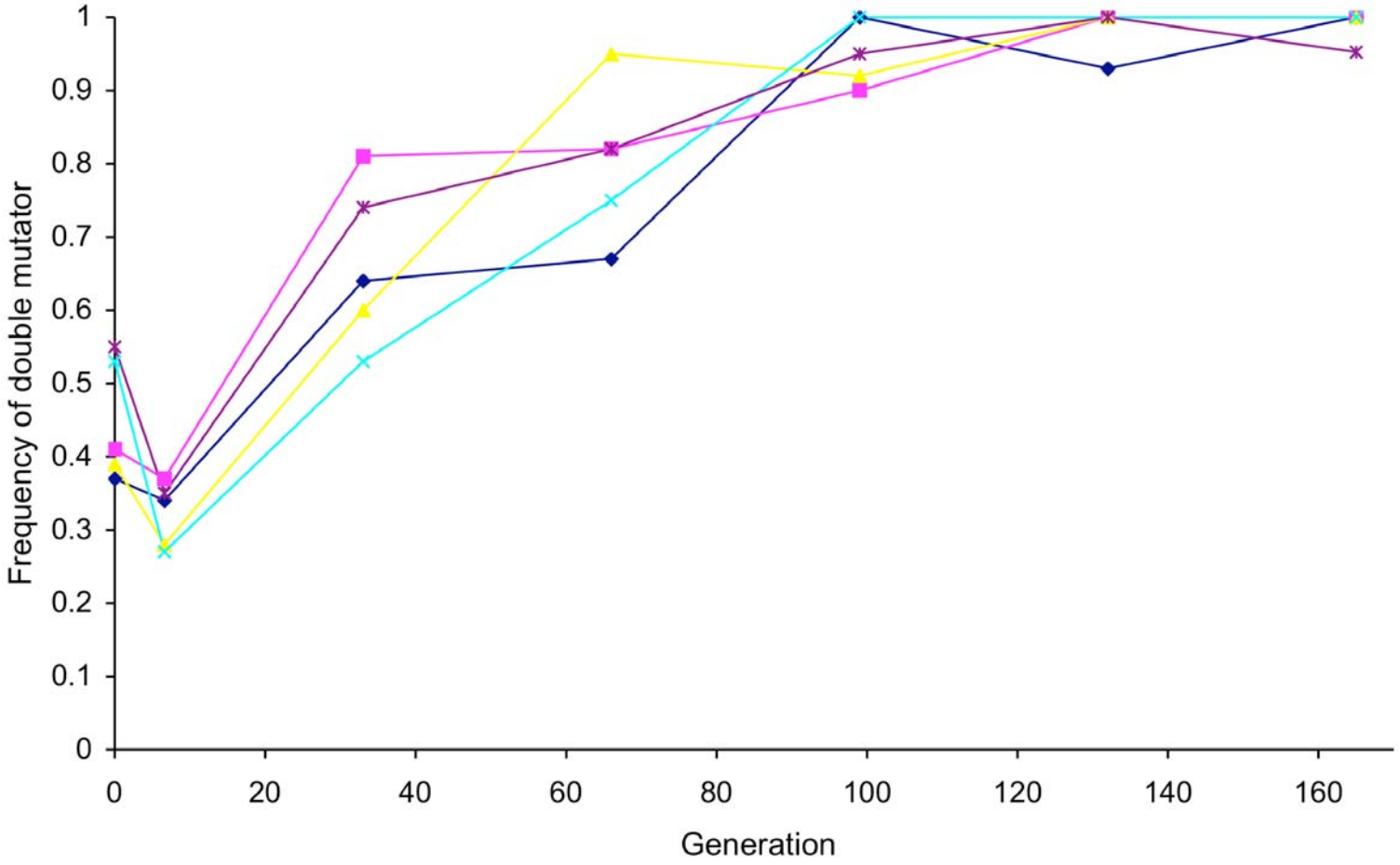


Figure 5. Mutation rates to nalidixic acid resistance measured in the constructed strains. Error bars give 95% confidence limits.

Note: Lynch and Kibota used a mutation accumulation assay to estimate $U_d > 0.0002$ for *E. coli* under experimental conditions similar to ours. Thus the 4,000-fold increase in mutation rate in our double-mutator strain suggests $U_d > 0.8$.

A double- vs single-mutator hitchhiking experiment



"Burndown" of double-mutator populations in ~1800 generations (!?)

Day 0:
Six separate populations of a double (2M) and a single (1M) mutator strain are established in minimal medium.
($N_e \approx 50{,}000$)

Everyday:
Each population diluted 1:1000000 and used to inoculate a fresh culture, giving about 20 generations of growth per day

Day 93:
2M population #2 goes extinct
Day 97:
2M population #6 goes extinct
Day 98: 2M populations #1,3,4,5 go extinct
All 1M populations extant

Preliminary assays suggest that fitness increased sharply early in the 2M populations, then declined sharply at the end. Mutation rate also appears to have increased greatly near the end, but more work is needed to confirm this.

"Conclusions"

- Adaptation by Darwinian selection is ultimately self-destructive.
- Most organisms in nature have to adapt now and then to survive changes in biotic and abiotic environments.
- Under Darwinian evolution, therefore, most organisms should have gone extinct.
- So...

"Conclusion"

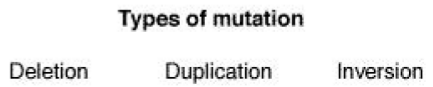
Darwin was wrong!!

Mutation

From CreationWiki, the encyclopedia of creation science

(Redirected from Mutations)

A **mutation** is any spontaneous heritable change in DNA sequence resulting from 2 possible mechanisms.

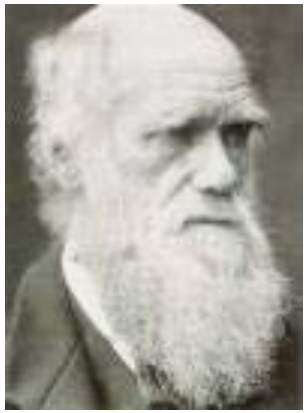


The paper released titled, *Complete genetic linkage can subvert natural selection* states that:

> *The key fact is that natural selection, although eminently robust, is a short-sighted process that favors traits with immediate fitness benefits. The fitness cost of mutator hitchhiking is generally not anticipated because of the slow accumulation of deleterious load. When a mutator hitchhikes with a new beneficial mutation, a simple model shows that the increased deleterious load due to the mutator is in fact suppressed during the spread of the beneficial mutation. Indeed, the full fitness cost of the mutator is only realized well after the beneficial mutation has stopped spreading. A mutator may therefore enjoy the immediate benefit of producing a new beneficial mutation without anticipating the eventual increase in deleterious load. Because of this delay in the accumulation of deleterious load, natural selection can drive mutation rate up to the point of no return.*
>
> ...
>
> *There is almost certainly no physiological barrier to such an effect in most organisms: the genomic mutation rate in organisms from viruses to eukaryotes is a quantitative trait affected by many mutations whose effects can readily cumulate to intolerable levels of error. In what follows, we show that there need not be a selective barrier to this process either: because the full fitness effect of increased deleterious mutation takes some time to accumulate after a higher mutation rate has evolved, it is theoretically possible for a population to evolve a critically high mutation rate and subsequently go extinct.*

"Conclusion"

Darwin was wrong!!



Further work: theory

- Extending the error threshold approach to incorporate finite, varying population size and effects of mutations on mutation rate itself.
- Exploring effects of recombination, diploidy.
- Extending simulations to very large (>1e6) population sizes.

Further work: experiments

- Detailed characterization of mutation rate, fitness and their covariance in existing "burndown" populations. (Relies on novel methods.)
- Further hitchhiking/extinction experiments at different population sizes, mutation rates.
- Sequence analysis of burndown populations.
- Incorporate recombination into system somehow?

Possible implications, if true, for

- Treatment of viral infections
- Age of asexual lineages
- Antiquity of recombination
- Genome structure

Many thanks to...

- **Alan Perelson**, Los Alamos National Lab
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- Szi-Chieh Yu, U. Illinois Chicago
- Chris Gentile, U. Pennsylvania
- Ruy Ribiero, Los Alamos National Lab
- Rob De Boer, Utrecht University
- Valeria Souza, Universidad Nacional Autonoma de Mexico
- *The Alfred P. Sloan Foundation, NSF, NIH, LANL, the UNAM, and FAPESP.*

Reference

PNAS

Complete genetic linkage can subvert natural selection

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Edited by Tomoko Ohta, National Institute of Genetics, Mishima, Japan, and approved February 16, 2007 (received for review August 22, 2006)

The intricate adjustment of organisms to their environment demonstrates the effectiveness of natural selection. But Darwin himself recognized that certain biological features could limit this effectiveness, features that generally reduce the efficiency of natural selection or yield suboptimal adaptation. Genetic linkage is known to be one such feature, and here we show theoretically that it can introduce a more sinister flaw: when there is complete linkage between loci affecting fitness and loci affecting mutation rate, positive natural selection and recurrent mutation can drive mutation rates in an adapting population to intolerable levels. We discuss potential implications of this finding for the early establishment of recombination, the evolutionary fate of asexual populations, and immunological clearance of clonal pathogens.

analytical models and simulations that support a different conclusion: under broadly defined conditions, the mutation rate can increase without bound in an adapting asexual population until it reaches an intolerable level that would presumably cause extinction. [Henceforth, all claims of extinction should be understood as being the inferred consequence of high mutation rates in conjunction with a rapid drop in fitness. See supporting information (SI) *Text* and refs. 11 and 12.] There is almost certainly no physiological barrier to such an effect in most organisms: the genomic mutation rate in organisms from viruses to eukaryotes is a quantitative trait affected by many mutations whose effects can readily cumulate to intolerable levels of error (1, 13). In what follows, we show that there need not be a selective barrier to this process either: because the full

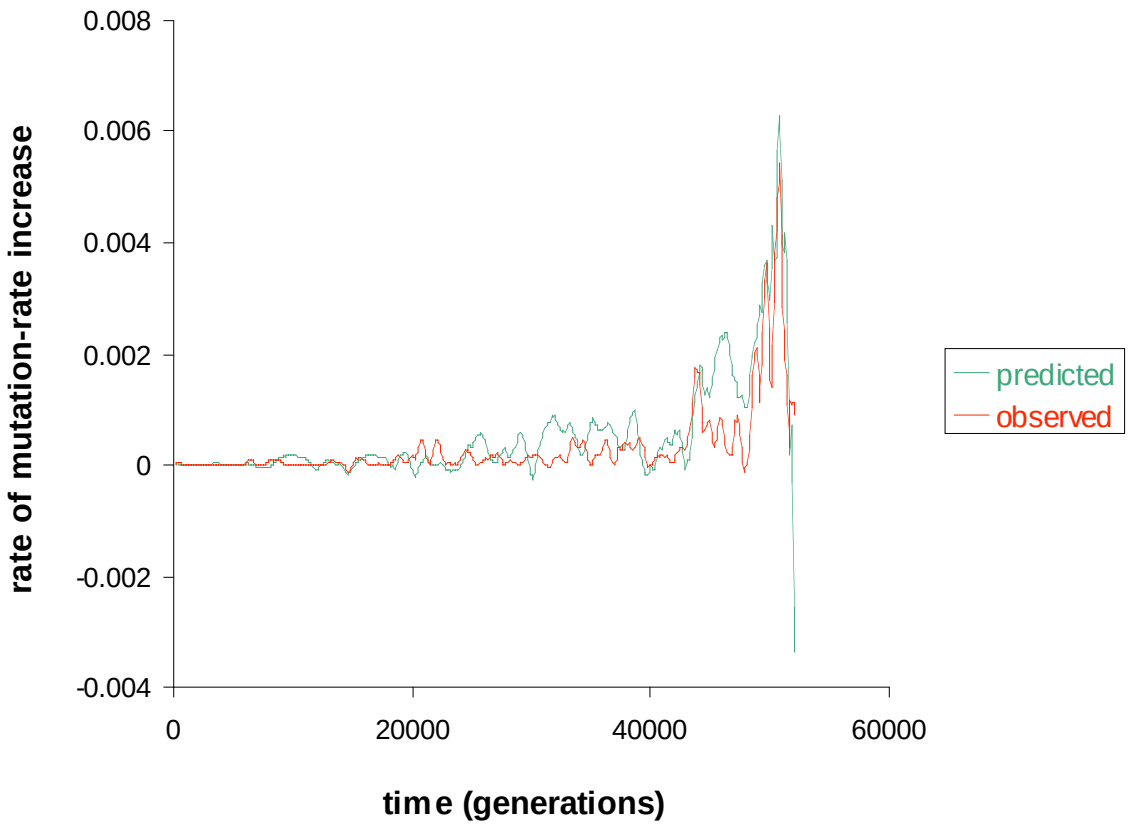
How it works - 1

Mutator hitchhiking

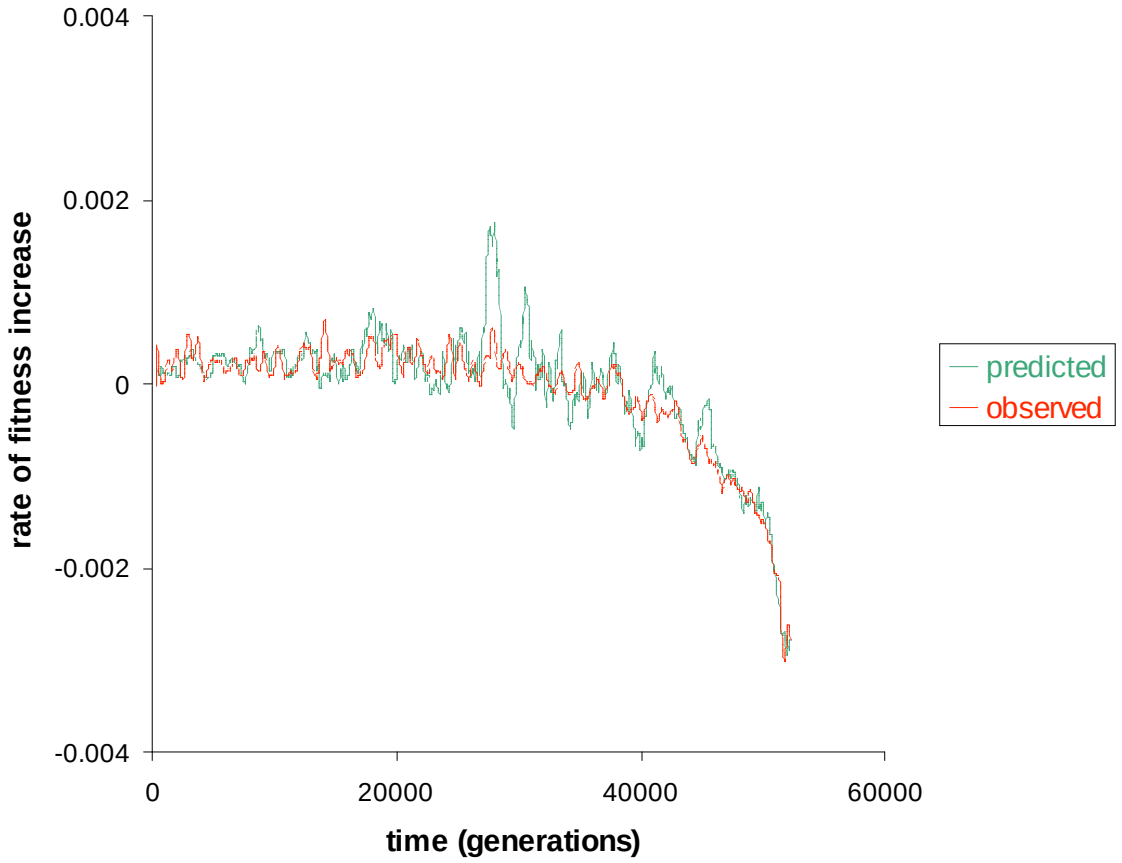
How it works - 2

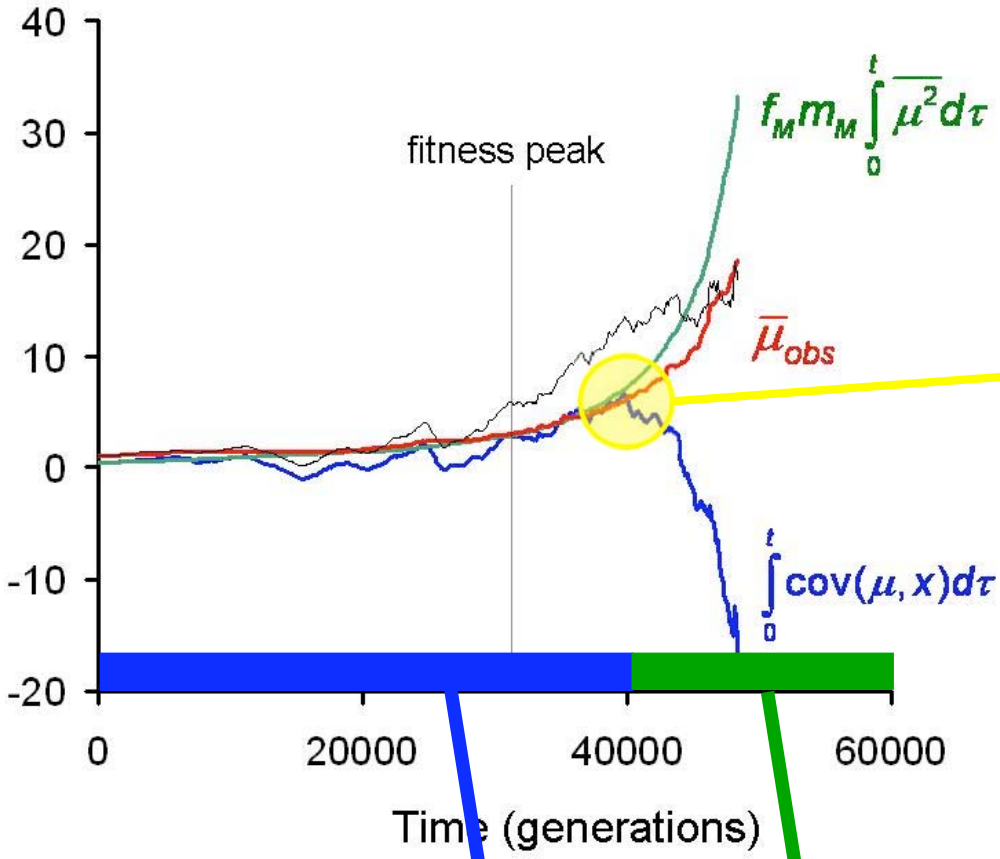
Mutation-rate ratchet

Analytical predictions



Analytical predictions





Hitchhiking dominates

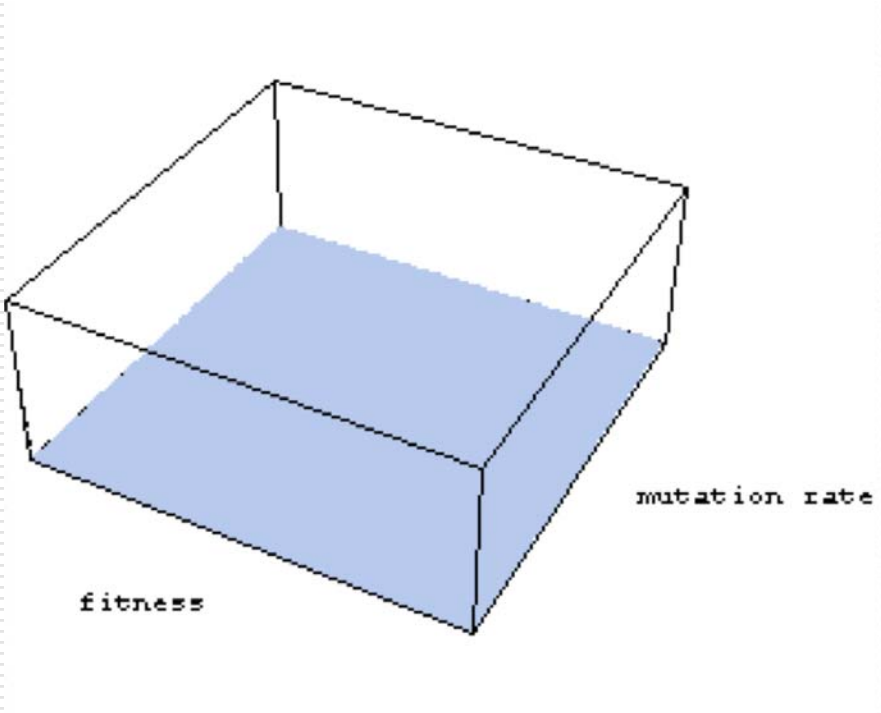
Generation ~40000

Mutation dominates

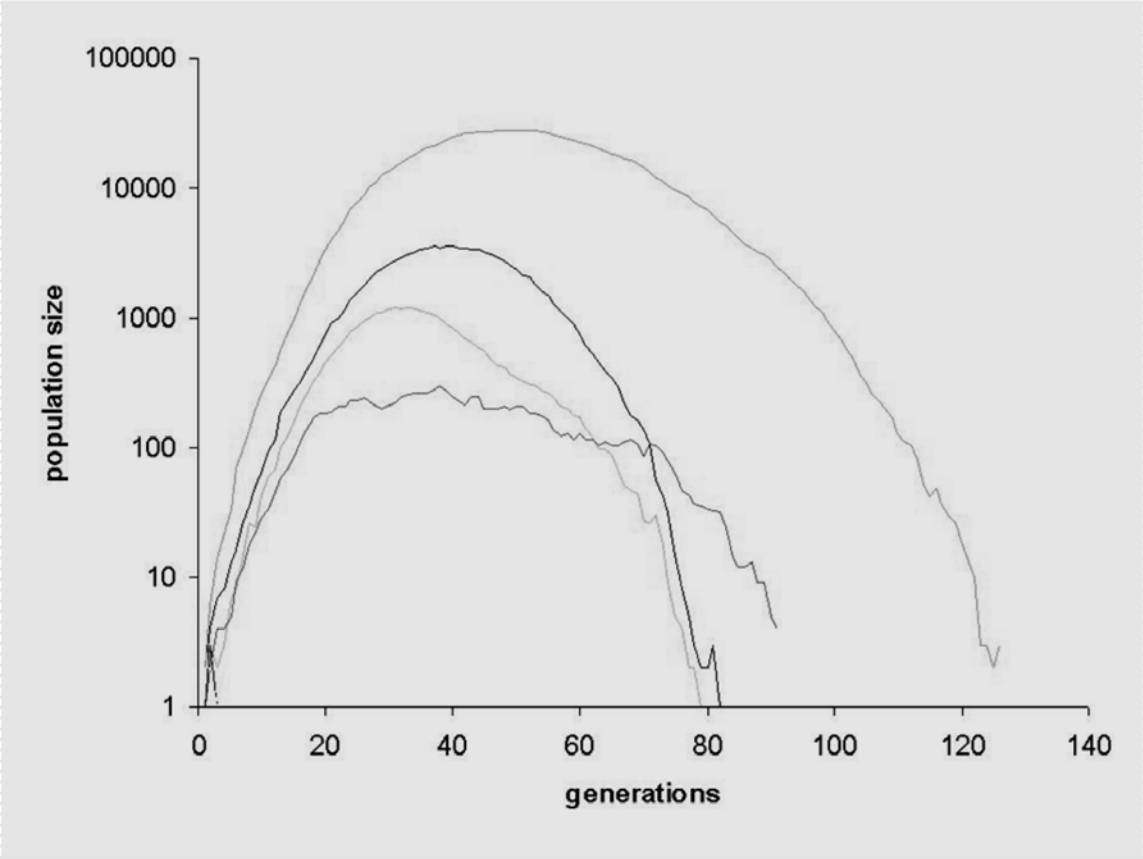
$$\partial \overline{\mu} / \partial t = \mathrm{cov}(\mu, x) + f_M m_M \overline{\mu^2}$$

Numerical solution

- 3D plot on an arithmetic scale.
- can't see the accumulation of mutators.
- but the sharpness of the "phase transition" is seen.



Growth of lineages in excess of fitness error-threshold



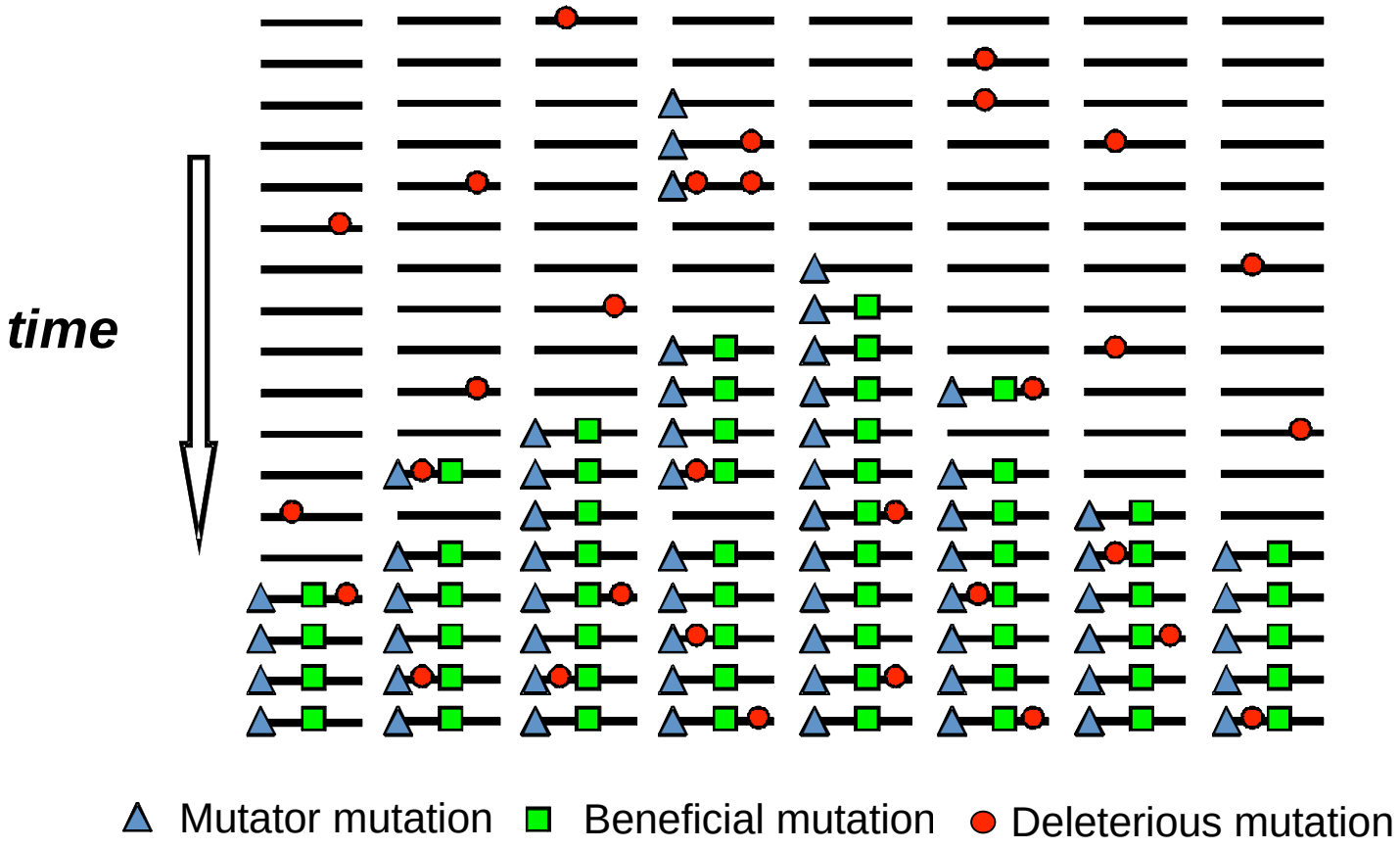
Mutators in natural populations

source	*frequency of mutators*
Jyssum, 1960	**~2% of samples of hospital *E. coli***
Gross and Siegel, 1981	**<1% of samples of coliform bacteria**
Suárez *et al.*, 1992	**7 of 60 samples of influenza virus**
Leclerc *et al.*, 1996	**~3% of samples from pathogenic populations of *E. coli* and *Salmonella enterica***
Matic *et al.*, 1997	**up to 15% of samples from commensal and pathogenic populations of *E. coli***
Oliver *et al.*, 2000	**20% of *Pseudomonas aeruginosa* isolates from cystic fibrosis patients**

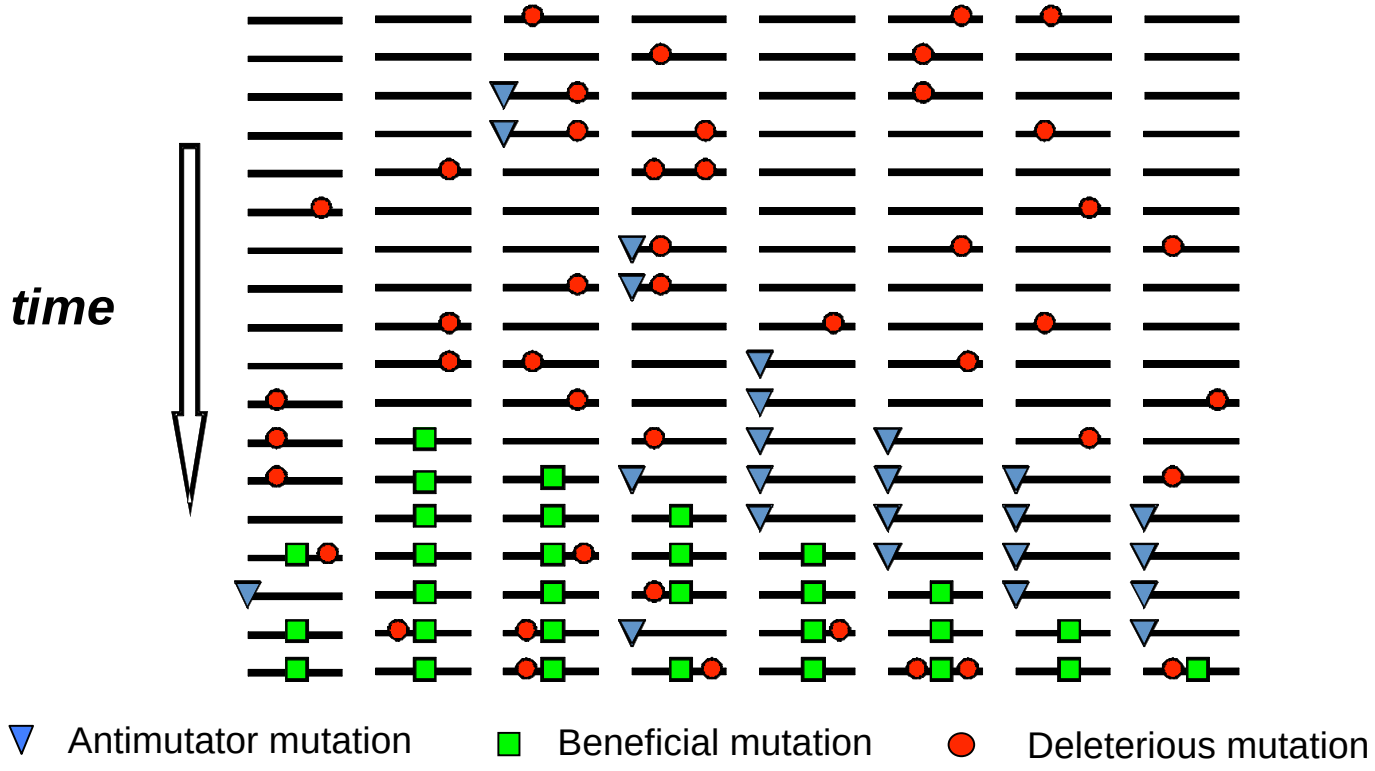
The CTG GCG repeat array is not well conserved

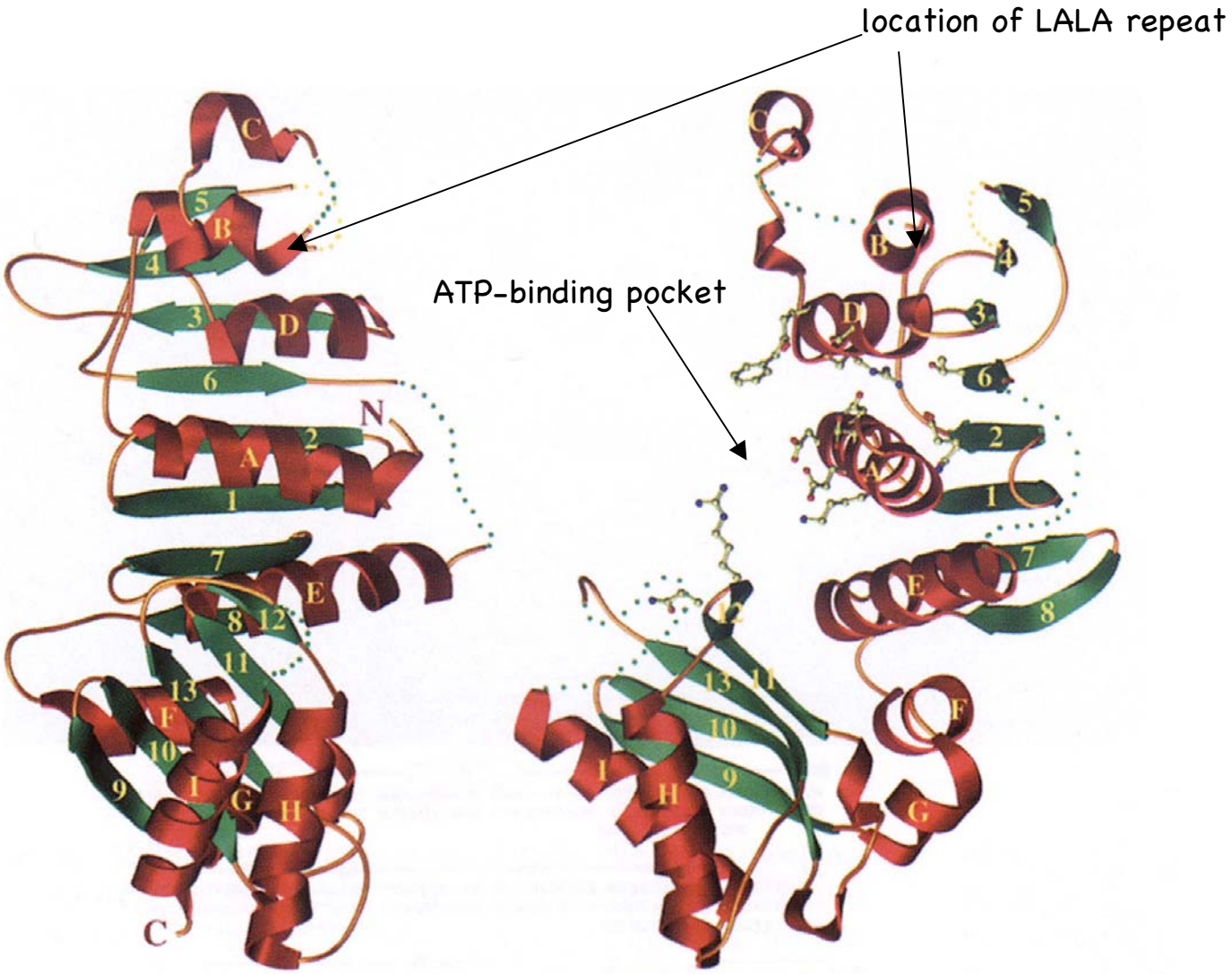
E. coli B	GAG CTG GCG CTG GCG CTG GCG CGT
E. coli O157:H7	... ...
E. coli K-12	... ...T ...
S. typhimurium	... ...C ...
S. enterica	... ...C ...
S. flexneri	... ...T ...
Y. pestis	..T T.. ..A ... ... T.. ..C ..C
P. aeruginosa	..C ... C.. ... ..C ... ..T ..C
B. subtilis	..T TGC AA. .GA ..T T.C CG. ..C
N. meningitidis	..C A.C .AA ..C ... ..C CAC ..C

Increase in population mutation rate through mutator hitchhiking



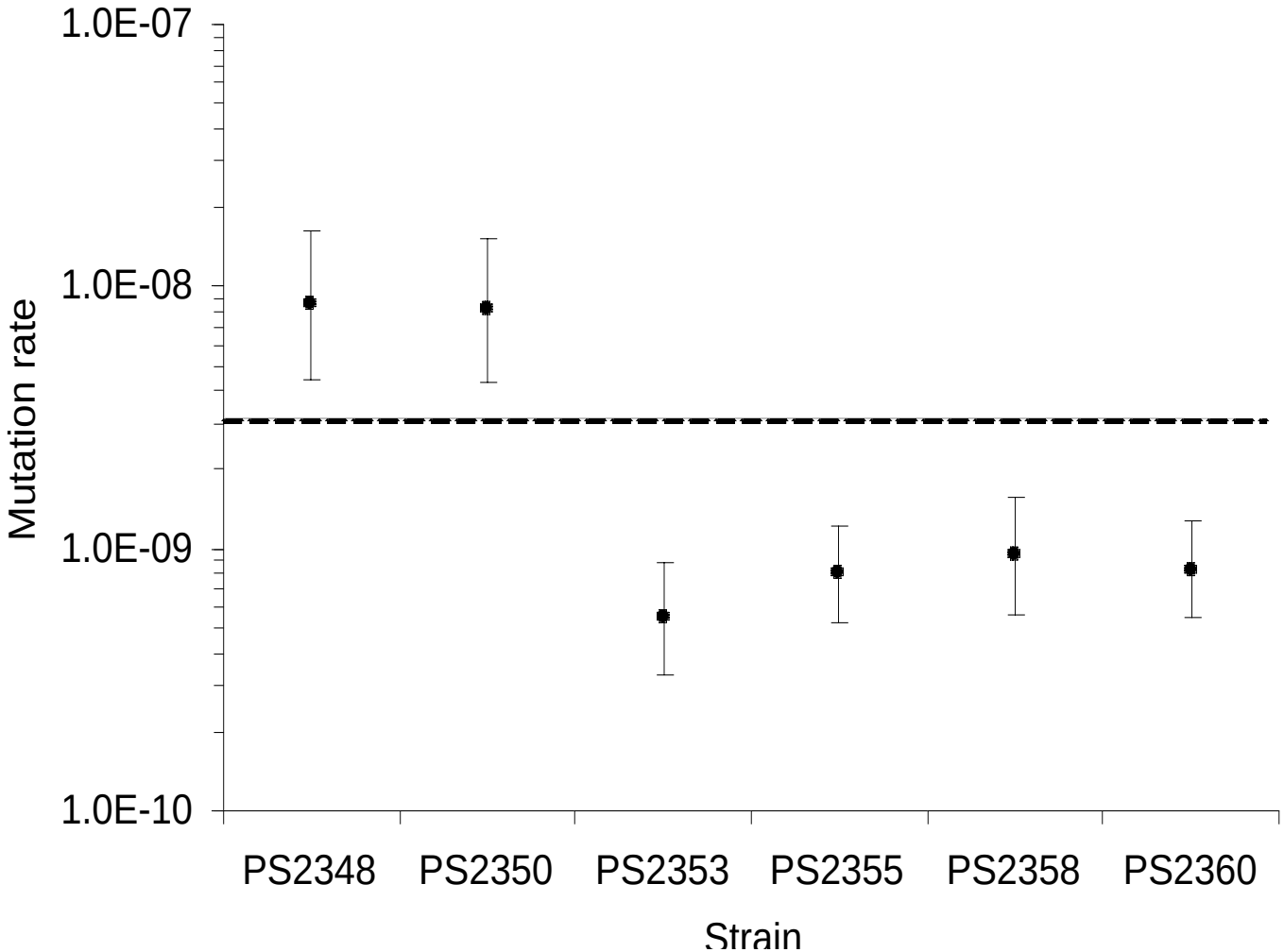
Failure to decrease population mutation rate due to anti-mutator's failure to hitchhike



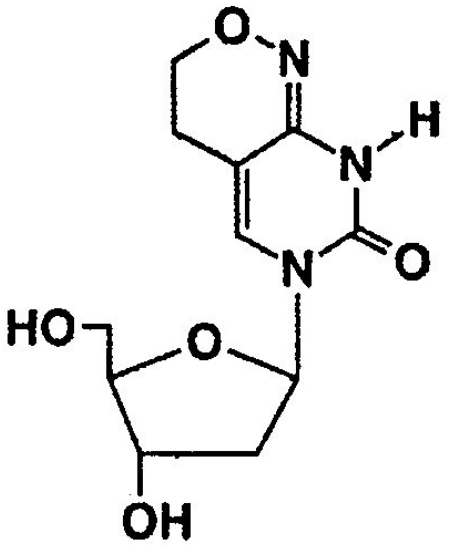


Source: Ban, C. and W. Yang. 1998. *Cell* 95, 541-552.

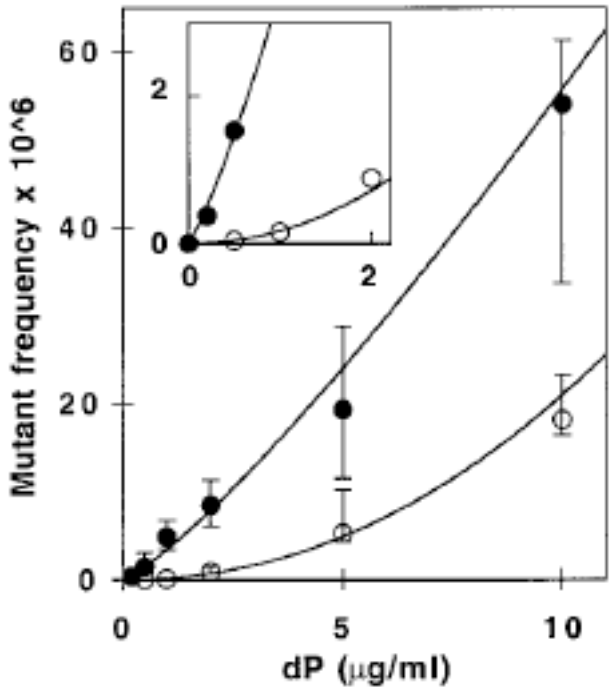
Allele replacement experiments confirm *mutL* repeat mutations as cause of mutator phenotype in Ara-2 and Ara-4 populations



Setting up an intolerable mutation rate (work in progress…)



deoxyribosyl-dihydropyrimido[4,5-*c*][1,2]oxazin-7-one (dP nucleoside): a powerful base analogue mutagen



Source: Negishi et al. 2002. Genetics 161: 1363-1371

Putting the hypothesis in context

- Many disadvantages of asexuality are indeed already known or have been hypothesized in theoretical work, but these are generally subtle evolutionary impairments that put asexual lineages at a disadvantage relative to their sexual counterpart. These subtle impairments do not present any real threat to an adapting asexual population by itself. In contrast to established theory, our findings would suggest that asexual systems are not only at a disadvantage when compared to sexual systems but that, *by themselves*, asexual systems are fundamentally flawed.

Link to classical theory

Fisher's fundamental theorem:

$$\partial \bar{x} / \partial t = \sigma_x^2$$

Modification due to linkage:

$$\partial \bar{x} / \partial t = \sigma_x^2 - f_D m_D \bar{\mu}$$

Link to classical theory

Price equation:

$$\partial \bar{\mu} / \partial t = \mathrm{cov}(\mu, x)$$

Modification due to linkage:

$$\partial \bar{\mu} / \partial t = \mathrm{cov}(\mu, x) + f_M m_M \overline{\mu^2}$$