- Rates of change from one state to another.

1) Progressive response to long-term directional selective challenges, e.g., global warming.

- How does the rate of adaptive evolution scale with population size?
- Which classes of mutations – small, medium, or large effects – contribute most to selection response?

2) Time to acquire a complex adaptation requiring multiple mutations to express a beneficial effect (epistasis), e.g., acquisition of antibiotic resistance.

- Are large populations more or less successful at establishing complex adaptations?

- Equilibrium distribution of mean phenotypes under constant population-genetic conditions.

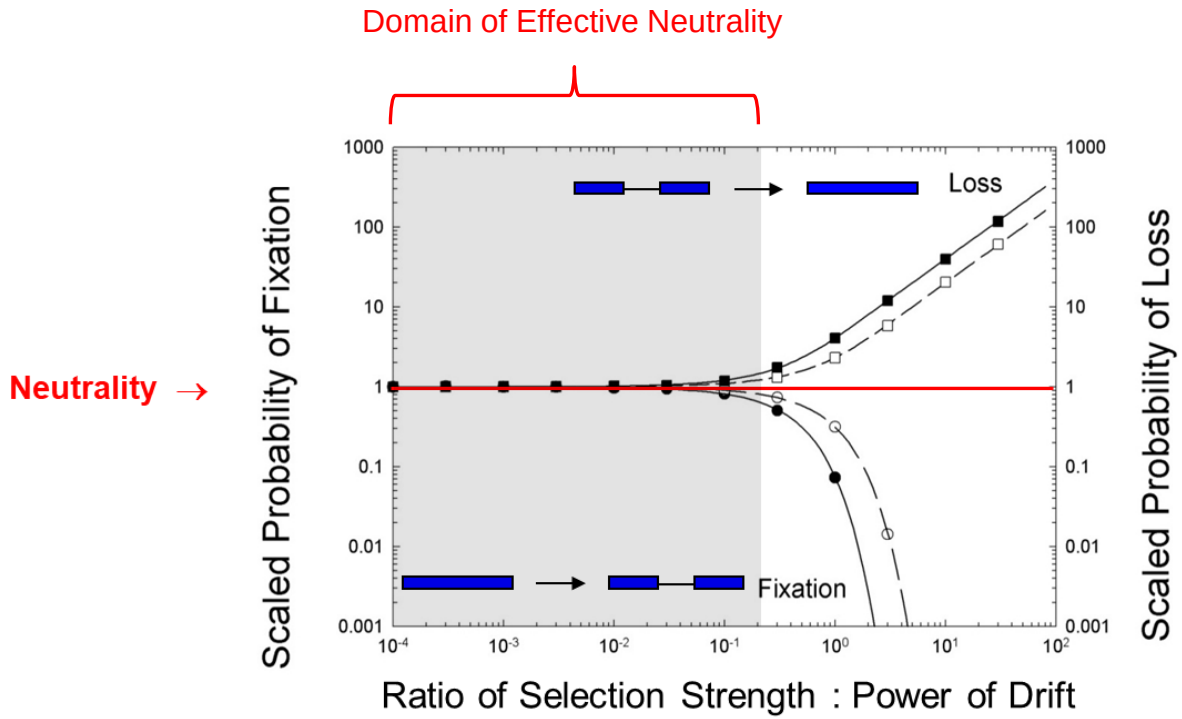
3) Steady-state distribution of mean phenotypes for traits under similar long-term selection pressures across the Tree of Life, e.g., enzyme efficiencies, replication fidelity, growth-rate potential.

- To what degree do mean phenotypes deviate from the optimum owing to the drift barrier and biased mutation?
- How far do mean phenotypes wander over time owing to the stochastic effects of mutation and drift?

The Importance of the Distribution of Fitness Effects of New Mutations

- Natural selection can only utilize mutations with fitness effects (s) larger than the power of drift ($1/N_e$).

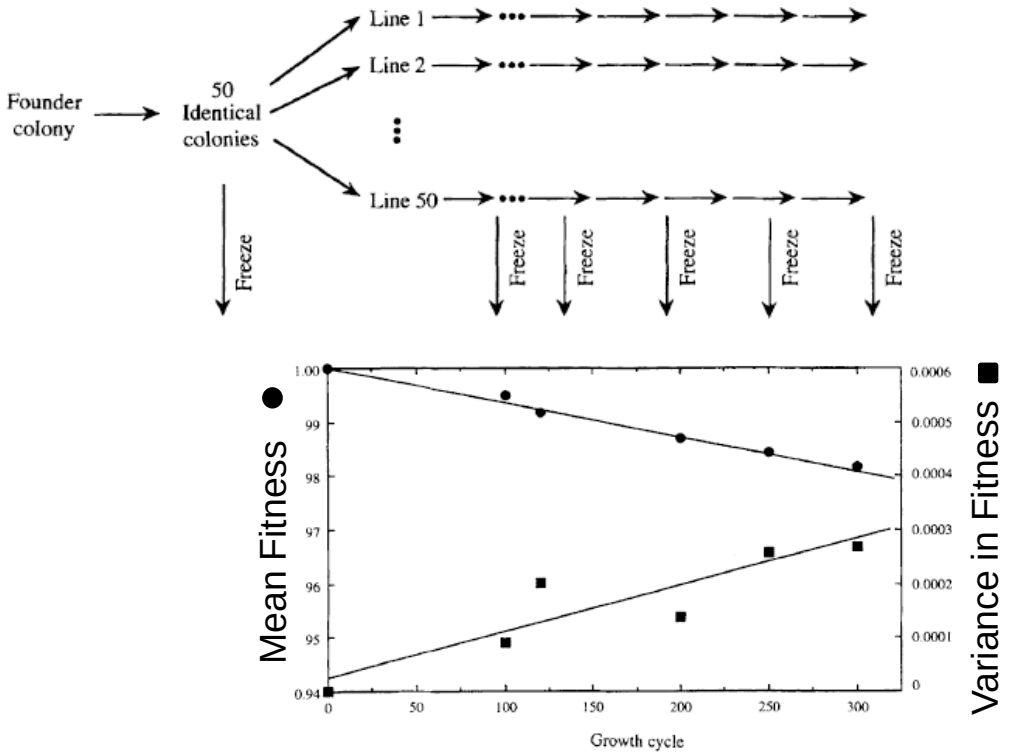
If the absolute value of $N_e s$ << 1, the probability of fixation is very close to the neutral expectation, and selection cannot prevent the passive establishment of a deleterious mutation or promote fixation of a beneficial mutation.



- For genetic drift to impose different barriers to the evolution of a trait in different lineages, there must be a substantial pool of mutations with small enough deleterious effects that they can drift to fixation in species with small but not large N_e.

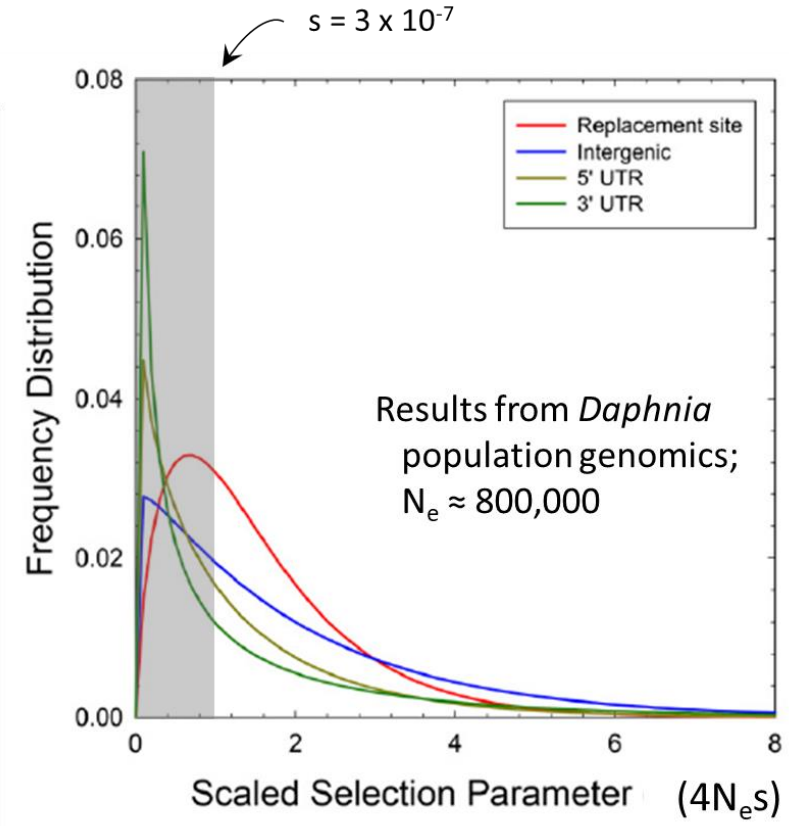
Multiple Lines of Evidence Suggest that Most Mutations are Deleterious and Have Small Effects

- Results from mutation-accumulation experiments invariably reveal slow declines in mean fitness, accompanied by increases in among-line variation.



Kibota and Lynch (1996)

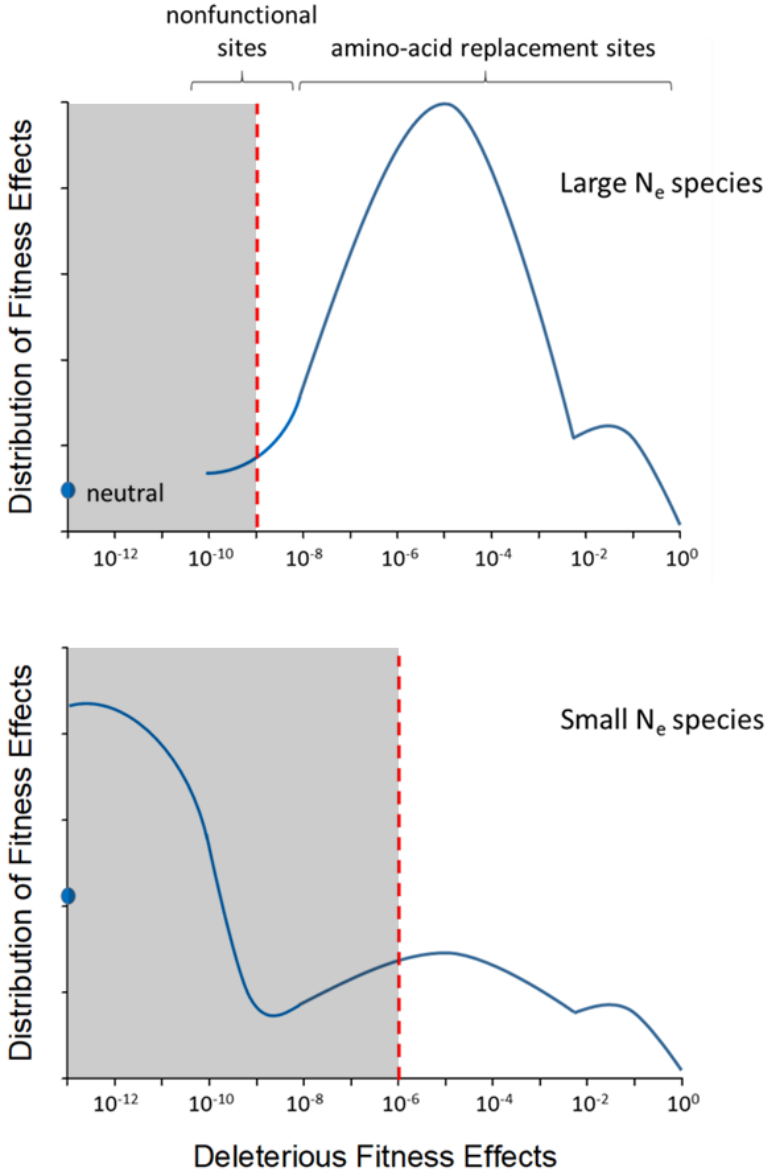
- Studies of the frequency distributions of mutations in natural populations can be used to indirectly infer the form of the distribution of deleterious effects.



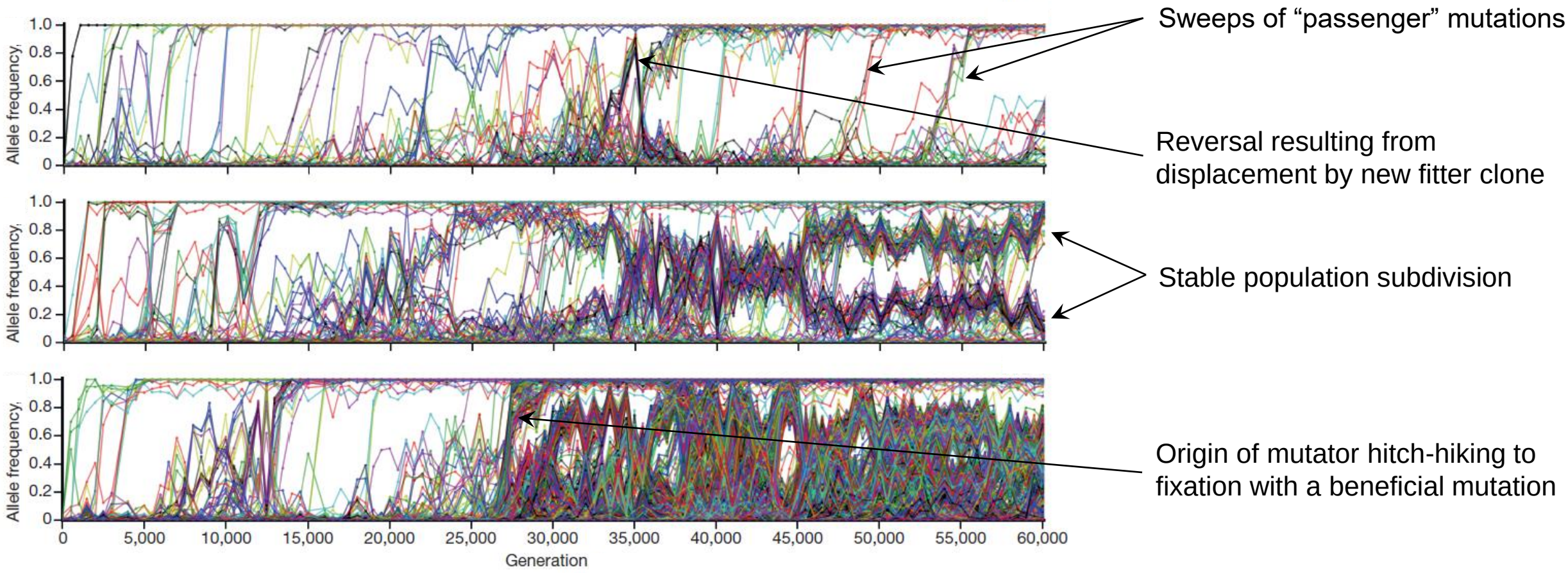
Lynch et al. (2017)

Bioenergetic Considerations: few mutations have absolute zero effects.

- The only fully neutral mutations may be A:T ↔ T:A and G:C ↔ C:G exchanges.
- The biosynthetic cost of a 1 base insertion ≈ 100 ATPs.
- The biosynthetic cost of an A:T pair is about 3 ATPs more than a G:C pair.
- The entire cost of building a bacterial cell is ~10^{11} ATPs.
- The cost of eukaryotic cells is 100- to 1000-fold higher.
- Minimum selective cost of a mutation in a bacterium is ~$3/10^{11} \approx 10^{-10}$.
- Minimum selective cost of a mutation in a eukaryote is ~$3/10^{14} \approx 10^{-13}$.



Observed Dynamics of Mutant Allele Frequencies in 10-mL *E. c*oli Cultures: 30% fitness increase over 60,000 generations.



10-mL cultures diluted 100-fold on a daily basis. To estimate allele frequencies, the complete genomes of each mixed culture were subject to pooled-population sequencing, to an average of 50x depth of coverage, every 500 generations over a 60,000-generation period. Each individual line in the plots denotes a mutation that arose to frequency 0.1 on at least one occasion. Results are shown for three replicate populations. All cultures were genetically identical and monomorphic at time zero. (Good et al. 2017).

1) The Classical Model of Sequential Fixation of Adaptive Mutations

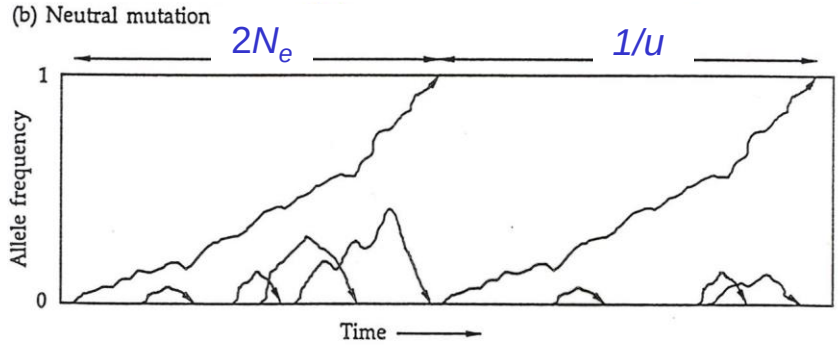
- The rate of adaptive evolution is equal to the product of the number of adaptive mutations arising per generation and the probability of fixation.

  N = absolute (haploid) population size
  N_e = effective population size
  u_b = rate of origin of beneficial mutations
  s = selective advantage

- Number of new mutations entering the population per generation = Nu_b

- Provided the selective advantage exceeds the power of drift, the probability of fixation $\approx 2s(N_e / N)$

- Long-term rate of fixation = $2N_e s u_b$



Neutral expectation:

Nu mutations arise per generation, and fix with probability 1/*N*.

Rate of evolution = (*Nu*) x (1/*N*) = mutation rate.

Areas of Uncertainty Regarding the Rate of Adaptive Evolution

- Ideal long-term rate of fixation = $2N_e s u_b$

All other things being equal, larger populations are expected to evolve more rapidly than smaller populations.

- Organisms with higher N_e evolve lower mutation rates, making the product $N_e u_b$ relatively constant?

- Smaller organisms with higher N_e have shorter generation times, which will increase the rate of evolution on an **absolute time scale**.

- Which mutational effects contribute most to the rate of fitness improvement: $s \times 2N_e s u_b = 2N_e s^2 u_b$
  - Mutations with small s may be more numerous, but s^2 declines rapidly with smaller s.
  - If $s << 1/N_e$, selection is ineffective.

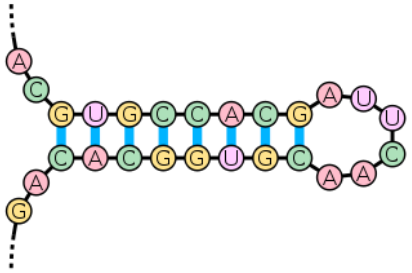
2) Vaulting Barriers to More Complex Adaptations

Evolution of a Complex Adaptation Through Deleterious Intermediates:

Haplotype:	**ab**	**Ab**	**aB**	**AB**
Fitness:	1	1 - s_d	1 - s_d	1 + s_b

Compensatory RNA stem-pair mutations:

A=U ⟷ G::U / A::C ⟷ G=C



“Compensatory pathogenic deviations” in sister taxa imply that evolution can proceed through deleterious intermediates

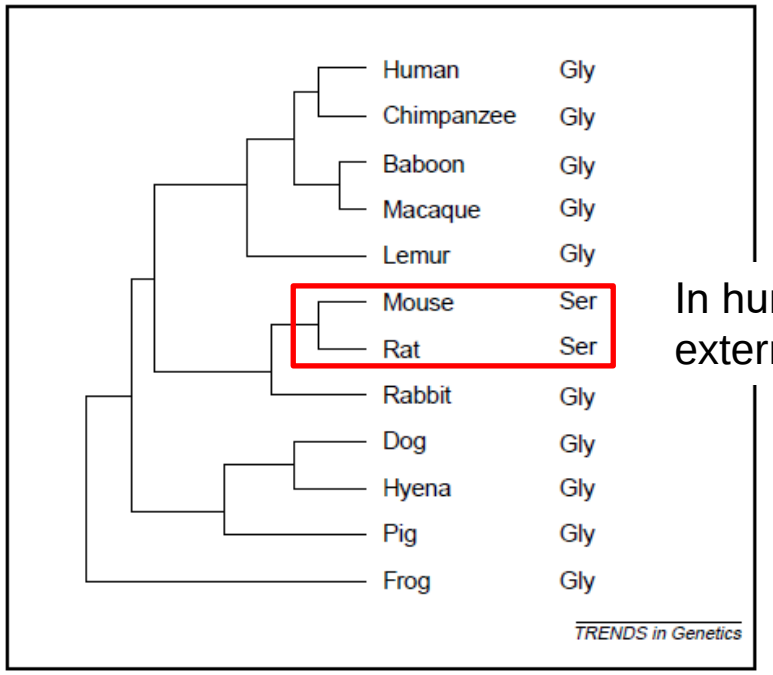


Figure 1. An example of fixed differences of disease-associated mutation (FDDAM). Amino acids at position 491 of the androgen receptors of 11 mammals and one non-mammalian vertebrate are shown. G491S leads to the complete androgen insensitivity syndrome in humans [2]. The tree topology follows [16] and the branches are not drawn to scale. The species names and GenBank accession numbers are: human (*Homo sapiens*) NP_000035; chimpanzee (*Pan troglodytes*), O97775; baboon (*Papio hamadryas*), O97960; macaque (*Macaca fascicularis*), O97952; lemur (*Eulemur fulvus*), O97776; mouse (*Mus musculus*), NP_038504; rat (*Rattus norvegicus*), AAA40734; rabbit (*Oryctolagus cuniculus*), P49699; dog (*Canis familiaris*), Q9TT90; hyena (*Crocuta crocuta*), AAM96904; pig (*Sus scrofa*), AAG40566; frog (*Xenopus laevis*), AAC97386. Abbreviation: Gly, glycine.

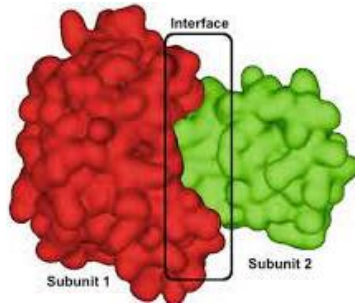
In humans, the Gly → Ser mutation in the androgen receptor gene causes males to have external female genitalia and other abnormalities.

- Many other examples of compensatory mutations in Kondrashov et al. (2002, PNAS, humans), and Kulathinal et al. (2004, Science, *Drosophila*).

Gao and Zhang (2003, Trends Genetics)

Cell Biology Provides Numerous Examples of Coevolving Sites Subject to Mutual Drift

- Most proteins assemble as multimers, requiring the coordination of specific residues on binding interfaces across proteins.



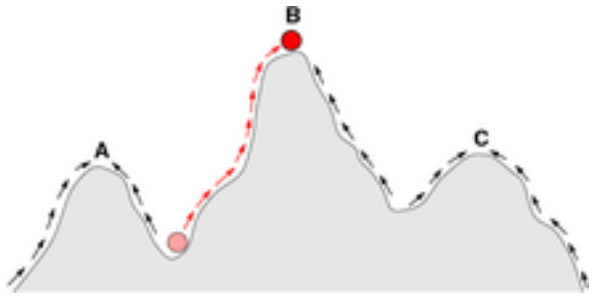
- For proper gene expression, recognition residues on transcription factors must closely match specific binding motifs on DNA.

- Signal transduction involves the relay of specific messages between receptors and response regulators via precise binding interactions.

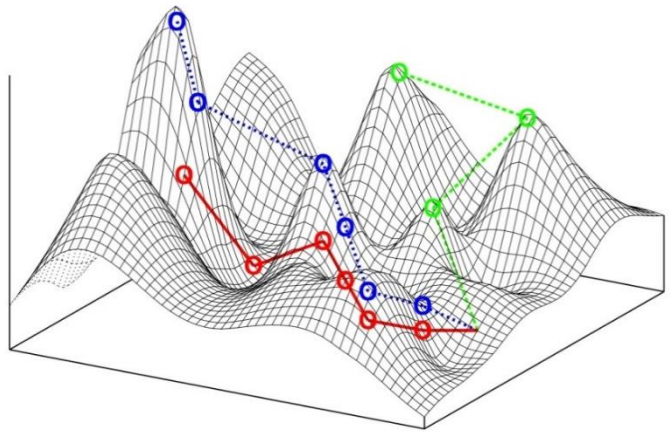
- Vesicle trafficking involves multiple layers of protein-protein interactions to achieve delivery of specific cargoes to appropriate locations.

The Adaptive Landscape: a Metaphor for Evolutionary Biology

- **A common but incorrect view**: selection cannot take a population from one adaptive peak to another unless the population size is small enough to allow maladaptive drift across the fitness valley.

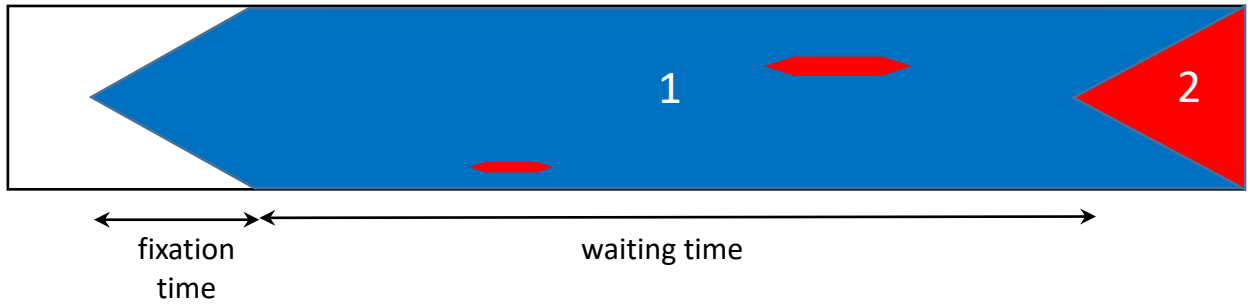


Phenotype →



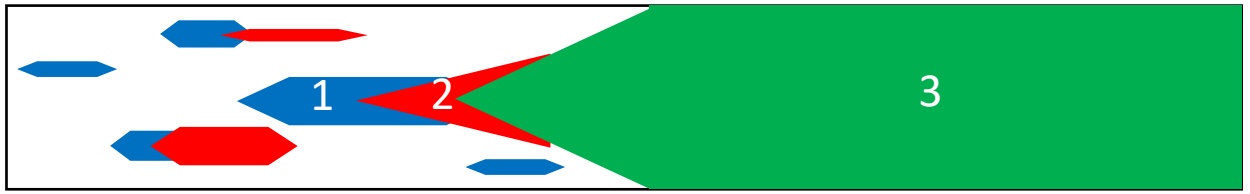
How do complex adaptations requiring more than one mutation become established?

Sequential fixation:



- In small populations, waiting times between mutations are long, and adaptation proceeds in a stepwise fashion, resulting in fixation of the intermediate state, and a sojourn through a mean-population fitness bottleneck.

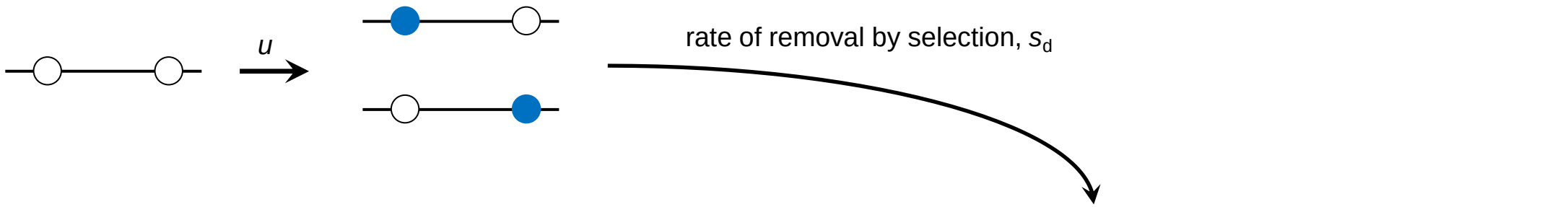
Stochastic tunneling:



- In larger populations, intermediate-state alleles need never be fixed, but are kept at low frequencies by selection-mutation balance, serving as launching pads for the final adaptation by double fixation.

Evolution by Compensatory Mutations: Deleterious Intermediates; Neutral End States; Diploid Population

Maintenance of intermediate-step deleterious alleles by selection-mutation balance:



Equilibrium frequency for each of the intermediate types = u / s_d

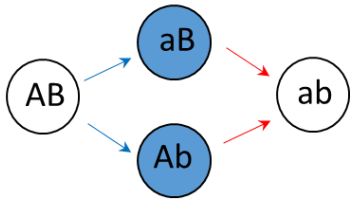
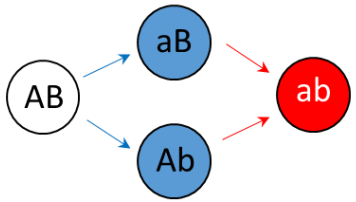
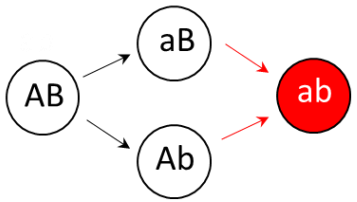
Equilibrium number of copies of intermediate alleles across both sites = 2 x $2N$ x u / s_d

x Rate of mutation to second-step alleles = u

x Probability of fixation of second-step allele = $1 / (2N)$

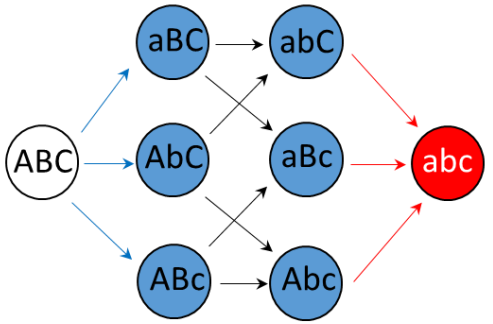
Rate of establishment of compensatory change = $(4Nu/s_d)$ x u / $(2N)$ = $2u^2 / s_d$, independent of population size.

The Likelihood of Alternative Paths of Evolution Can Be Strongly Modulated by Changes in Population Size

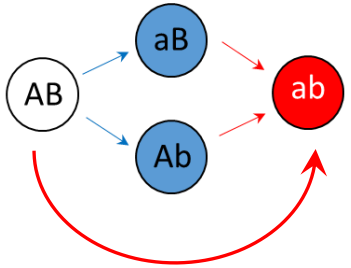
	Per-generation Rate of Transition from AB to ab	Scaling with N_e Assuming u is Inversely Related to N_e
Neutral endpoints, deleterious intermediates: 	$2u^2 / s_d$	proportional to $(1/N_e)^2$
Beneficial endpoint, deleterious intermediates: 	$8N_e u^2 s_b / s_d$	proportional to $(1/N_e)$
Beneficial endpoint, neutral intermediates: 	$4N_e u s_b$	independent of N_e

Alternative Paths to the Final Advantageous State

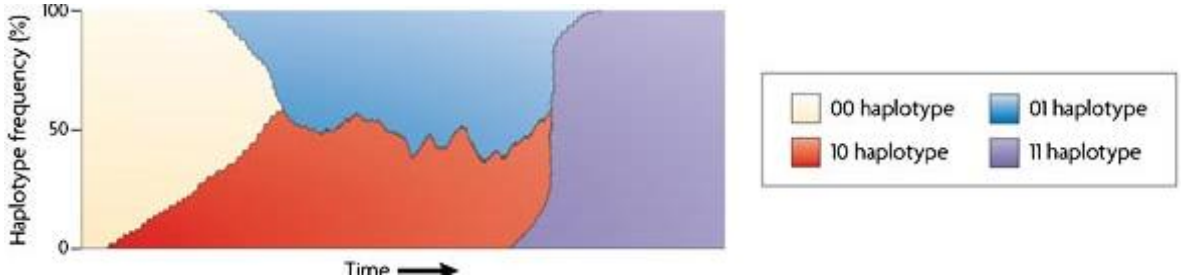
- Additional intermediate states can prolong things, but in some cases the rate of transition scales with no more than the square of the mutation rate.



- Multi-nucleotide mutations are common, and can create the final adaptation in a single event, by-passing the deleterious intermediate.



- Recombination need not enhance the rate of establishment.
  - Serves as a complete barrier to transition if the rate exceeds the final selective advantage.
  - Optimal rate is context dependent, ≈ s/2.

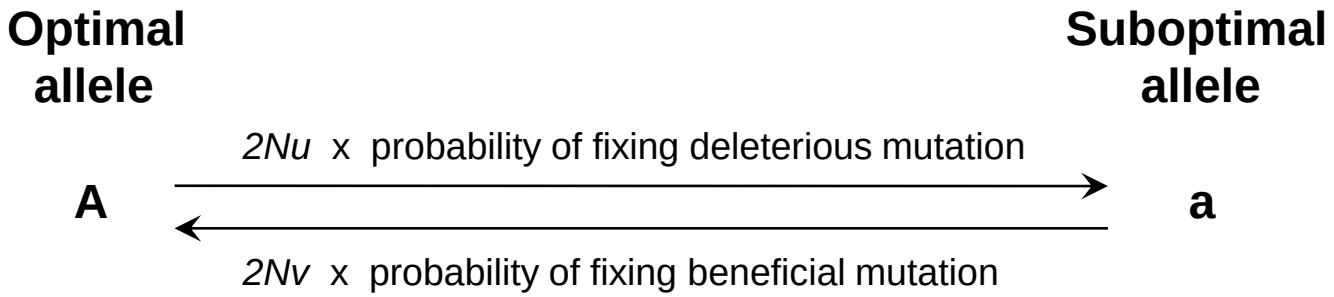


3) The Phylogenetic Dispersion of Mean Phenotypes

- Many cellular traits have retained the same function for hundreds of millions of years, and may have been under nearly invariant selection pressures for this same amount of time.

- This shifts the evolutionary focus away from dynamical changes in allele frequency under directional selection to the long-term steady-state probability distribution of alternative phenotypic states.

Detailed Balance: the long-term equilibrium distribution of alternative population states.



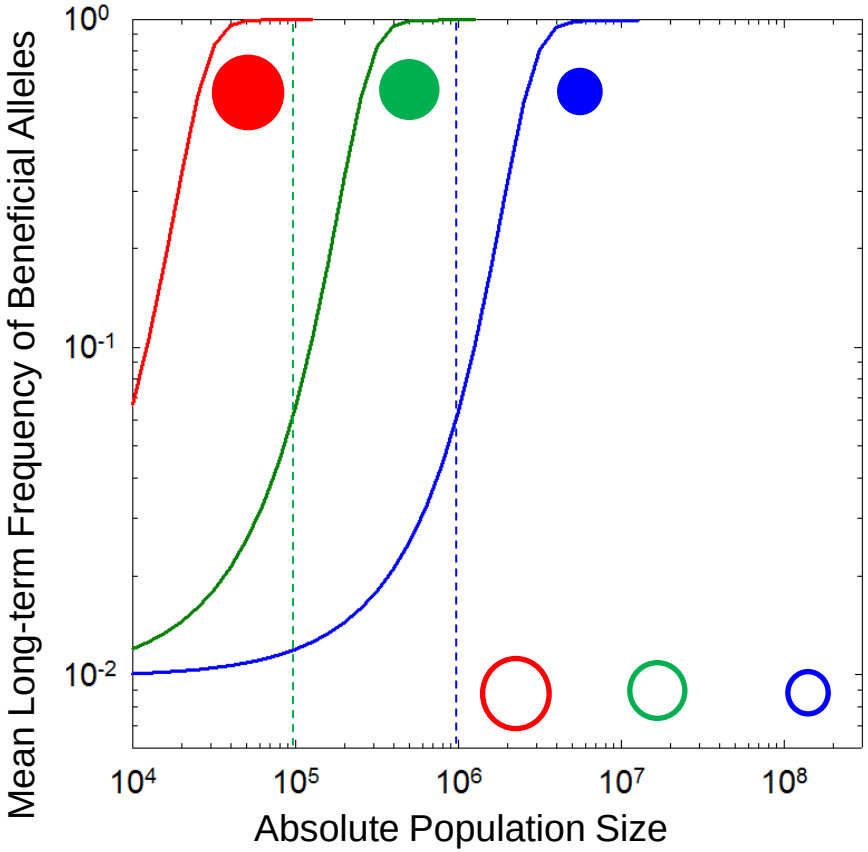
- N = the number of individuals in the diploid population.
- u and v = mutation rates to deleterious and advantageous alleles, so $\beta = v/u$ is the mutation bias towards the favorable allele.
- Ratio of fixation probability for a favorable relative to a deleterious allele (the selection bias) is e^S, where $S = 4N_e s$.

At equilibrium, the rates of transition in both directions must be equal: the deleterious state is rare, but when it occurs, it rapidly transitions back to the beneficial state.

Probability of being fixed for the deleterious allele $\simeq \frac{1}{1+\beta e^S}$ ← net pressure to the favorable allele from mutation and selection pressure

Expected Frequencies of Fitness-Improving Alleles:

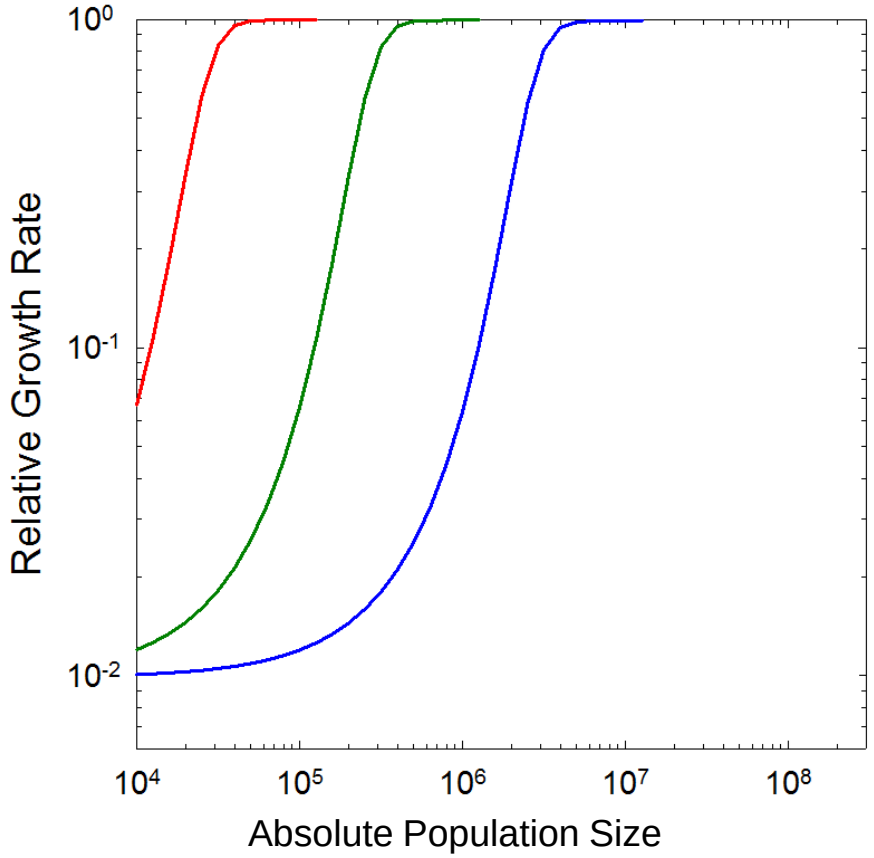
S = 10^{-4} 10^{-5} 10^{-6}

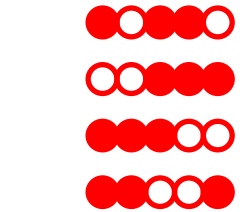


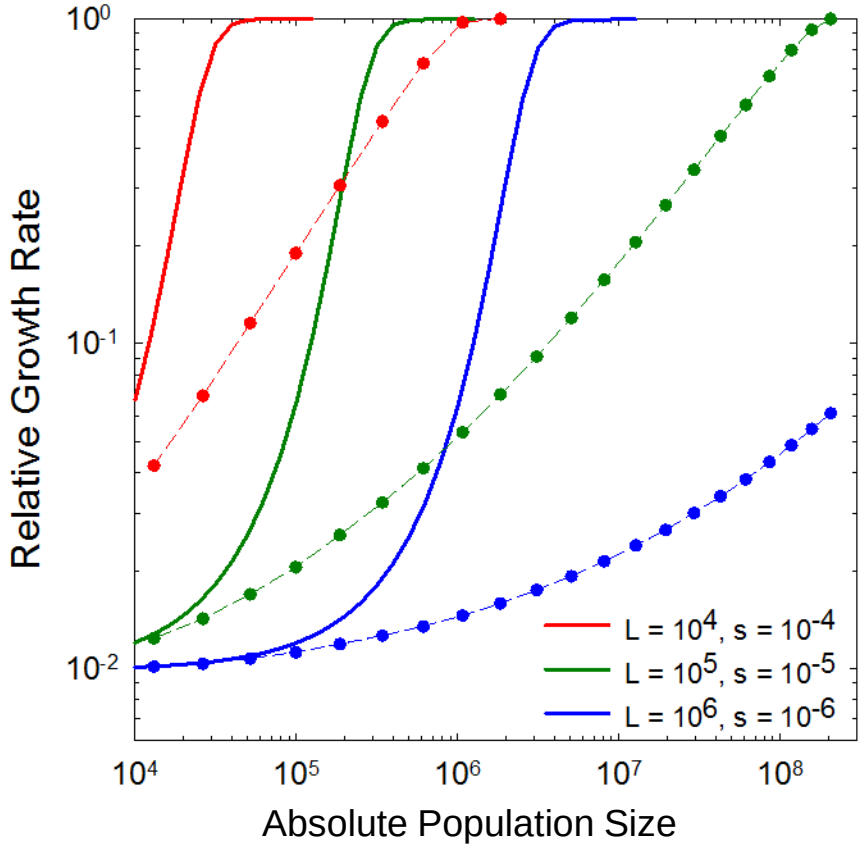
- Gradient is steep with freely recombining loci – inflection is at point where the power of selection ≈ power of drift; $s \approx 1/N$ or $Ns \approx 1$.

A simple shift to linkage blocks greatly flattens the gradient:

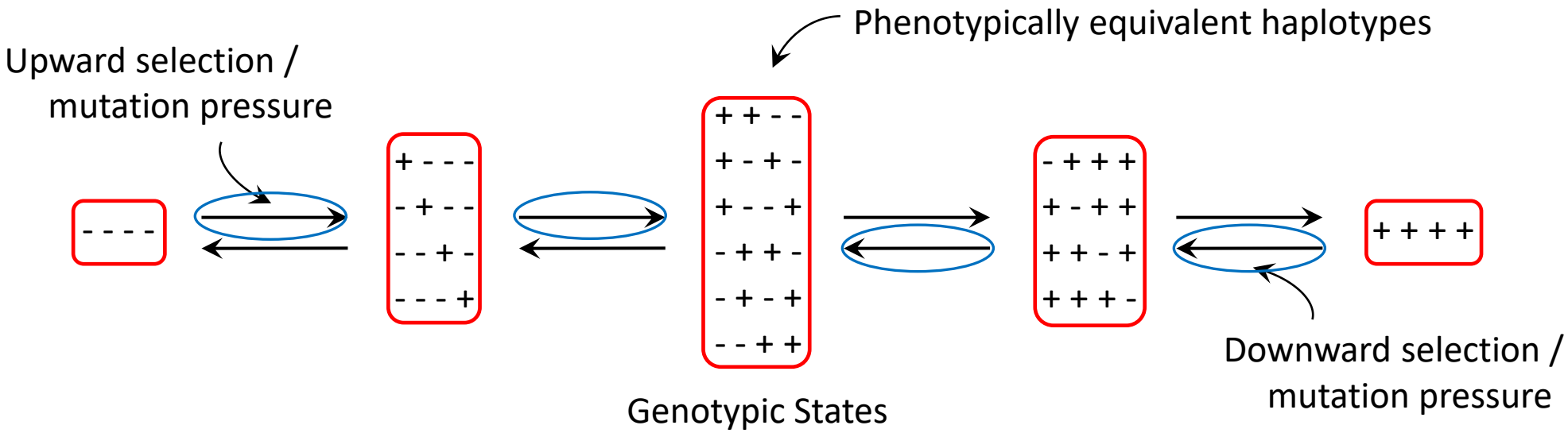
- Population behaves genetically as though it is much smaller than the census size.







Extension to Multilocus Traits, e.g., matches along a transcription-factor binding site



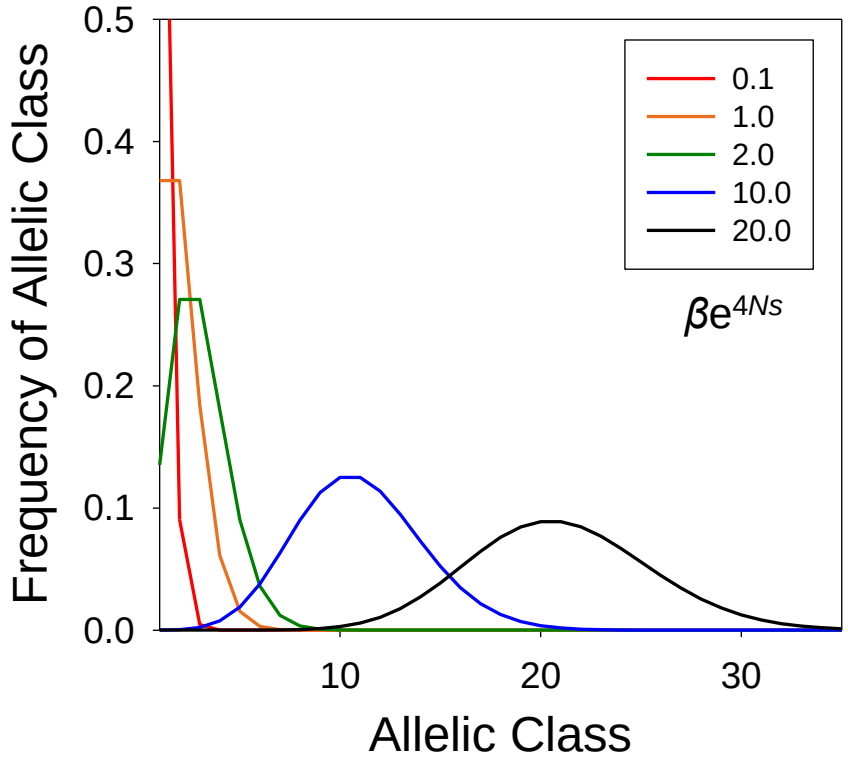
Equilibrium frequency = product of net pressures on arrows pointing towards state from both directions.

The Steady-state Evolutionary Distribution

- The probability distribution of alternative states is Poisson, with key parameter βe^{4Ns}.

- Substantial phenotypic variation among lineages, **even when selection and mutation operate identically in all lineages.**

- Most common state is not necessarily the optimum.



- Under effective neutrality (Ns ≈ 0), the form of the distribution is independent of population size.

Summary

- Despite the common view that populations under identical selection pressures will tend to be highly similarly phenotypically, many plausible situations exist in which uniform selection combined with random genetic drift and/or mutation bias can lead to substantial interspecies divergence, sometimes more than expected under drift alone.

- Raises questions about the common assumption that observed mean phenotypes provide a perfect reflection of the optimum defined by prevailing selection pressures.

- The drift-barrier hypothesis predicts that the mean phenotypes of some traits will exhibit gradients, with the level of functional refinement increasing with N_e.

- Mutation bias can impose evolutionary attraction towards a particular region of phenotypic space, in ways that may conflict with or reinforce prevailing selection pressures.

- Even if unbiased, mutation influences the expected distribution of mean phenotypes because genotypic states differ in the multiplicity of ways in which they can be constructed from the underlying set of genetic loci.