Small Populations: Inbreeding

Inbreeding

- Mating between related individuals
  - Individual instances
    - generally outbreeding population; one-off matings of related individuals
  - Regular systems of inbreeding
    - e.g. creating recombinant inbred lines
  - Local breeding structures
    - e.g. based on proximity; assortative mating
  - Overall relatedness within small populations

Common misperception

- Inbreeding leads to departures from Hardy-Weinberg equilibrium genotype frequencies.
  - Not necessarily true.
  - (And: departures from H-W genotype frequencies lead to an excess of homozygotes, uncovering rare recessive alleles. This can be true, but not necessarily a function of inbreeding *per se*).

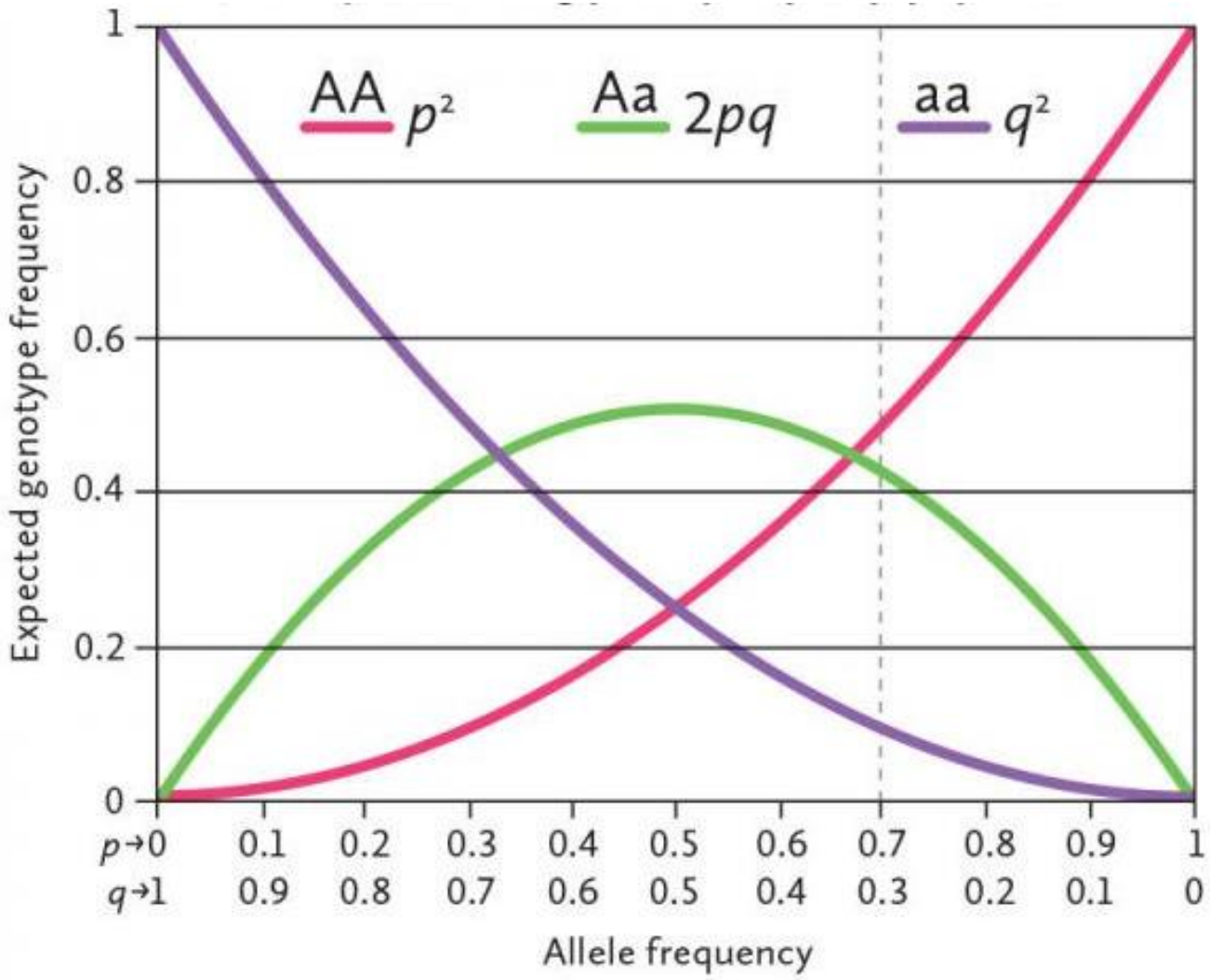
The actual problem

- In smaller populations, drift is a stronger force than selection – deleterious alleles can increase in frequency.

* The frequency of a homozygous genotype increases as the allele frequency increases.
  - Recessive deleterious alleles are uncovered.

- Also … allele frequencies depend on population sizes.
  - If a population contains 20 diploid individuals, the rarest allele has a frequency of 1/40.

H-W genotype frequencies



Small population sizes

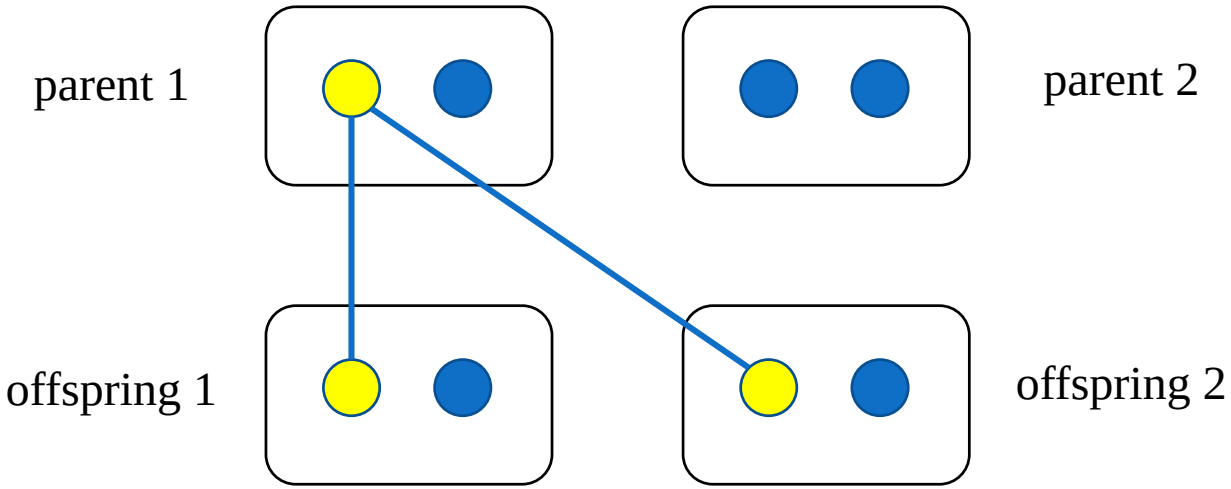
- Rare alleles can become common via drift.
- Deleterious homozygous genotypes become more likely.

- Increase in relatedness between individuals is also a consequence of small population sizes.

Inbreeding

- Individuals in the population tend to carry more and more alleles that are **identical by descent** (IBD).

Identity by Descent (IBD)



- Alleles that derive from a common ancestral allele are IBD.

Inbreeding Coefficient

- Measure inbreeding via the **Inbreeding Coefficient**:

  F_t = Pr (2 alleles w/in an individual at a locus are IBD)

Inbreeding coefficient in gen t+1

- Generating inbreeding: F_{t+1}

  Sample the first allele, then …

  - this allele is sampled again (new inbreeding in gen t+1)

    or

  - a second allele is sampled, but it was already IBD with the first allele at gen t (old inbreeding)

Inbreeding coefficient in gen t+1

- $F_{t+1} = \frac{1}{2N} + (1 - \frac{1}{2N})F_t$

new inbreeding

old inbreeding

N = population size (# individuals)

- If $F_0 = 0$, then:

$$F_t = 1 - (1 - \frac{1}{2N})^t$$

Inbreeding Coefficient

- Increases over time as alleles in a population are lost to drift.

- Eventually, one allele will become fixed in a population …
- F_t = Pr (2 alleles w/in an individual at a locus are IBD) $\Rightarrow$ 1.

Inbreeding exercise

- statgen.ncsu.edu/dahlia/inbreeding
  - [click on ‘go’]
- Simulates a small population over time.
- First value in each row is the generation number, starting at zero.
- Next are the genotypes of the thirteen individuals in the population.
  - [how many alleles are there at this locus?]

- Successive generations are simulated until only two alleles are left in the pop.
  - How many generations did this take?
  - Will this be the same every time the simulation is run? Why or why not?
- What are your expectations for the inbreeding coefficient of this population at this point?
- What are your expectations for genotype frequencies at this point?
  - Do you expect H-W genotype frequencies in this population?

- What information does the second-to-last column (next to the genos) provide?
  - Can this number increase between generations? Why or why not?
- What information does the final column provide?
  - Does this value always decrease over time? Why or why not?
- At the bottom of the page, a χ^2 statistic is given. The null hypothesis tested is: "H_0: genotype frequencies follow H-W expectations." What results do you get?

Some take-home messages

- Small populations are affected strongly by drift.
- Alleles will be lost over time.
  - Which alleles are lost is random.
- The inbreeding coefficient increases over time as alleles are lost.
- We still may expect to find H-W genotype frequencies in the population.

* Genetic variation is reduced over time.

Inbreeding Depression

Fragmented Populations

Fragmented populations

- Large natural population with gene flow across the population is fragmented into a number of smaller populations.
  - Habitat destruction; creation of man-made barriers; alteration of landscapes.

- What are the consequences?

Fragmented populations

- The smaller subpopulations are more susceptible to the forces of drift …
  - drift acts more quickly within smaller sub-populations than it did in the larger original population.

- Variation will be lost in subpopulations.
- Sub-populations will start to diverge.

Fragmentation exercise

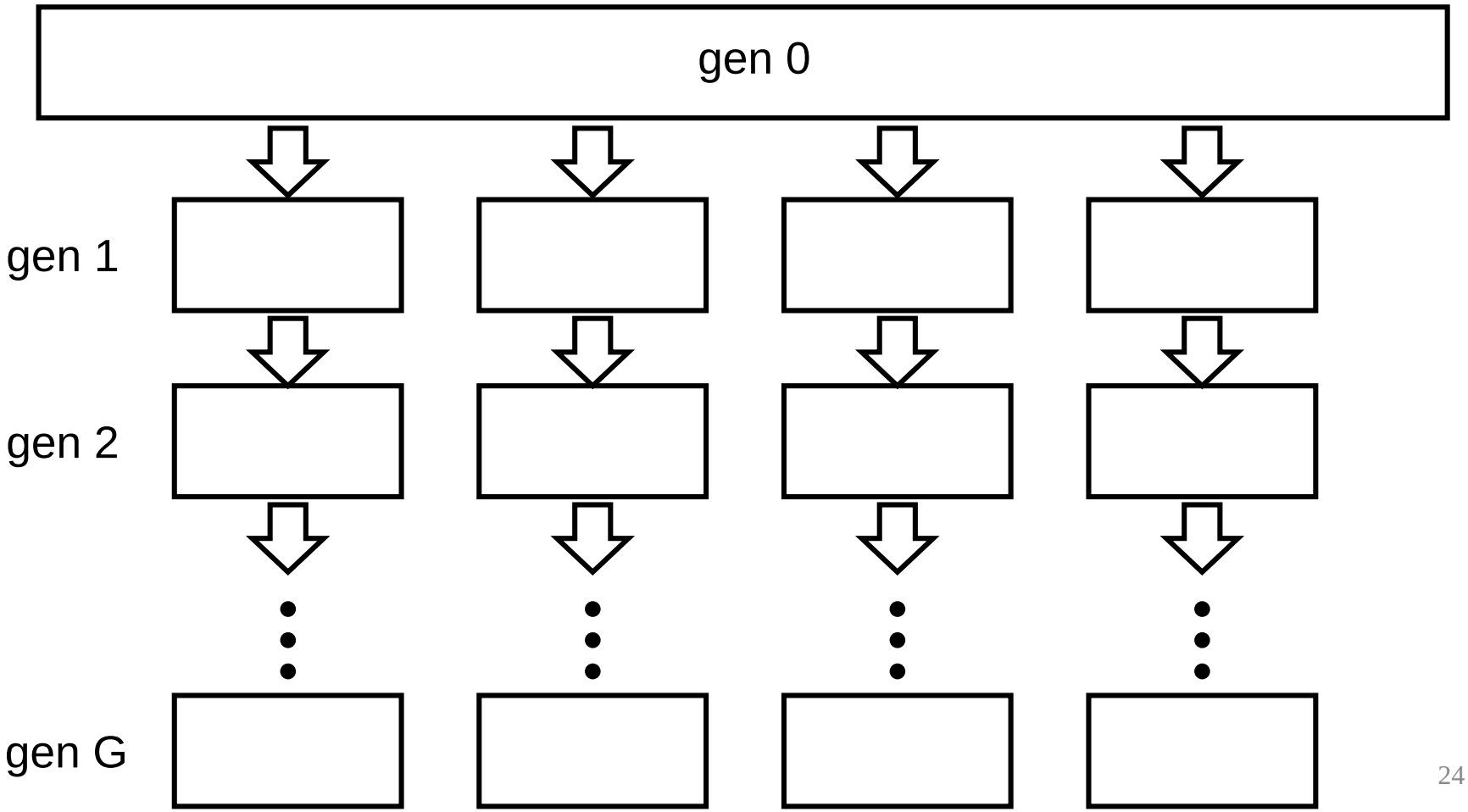
- Can we measure the amount of divergence between subpopulations?

- Run the inbreeding tool a few (say 5-6) times.
- Consider each simulation to be one subpopulation from a historically larger, now fragmented population.

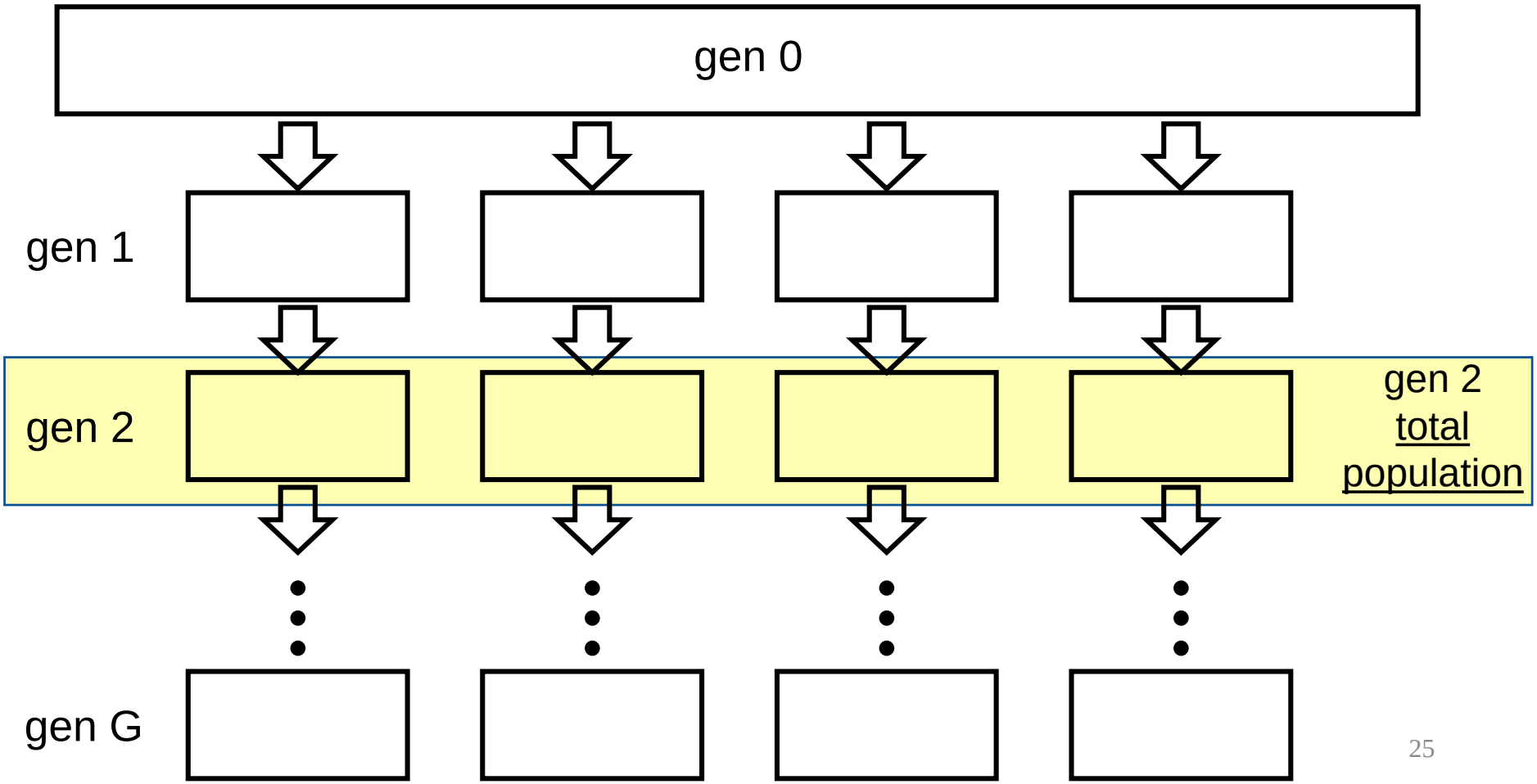
Exercise

- Extreme case: no migration between subpopulations.
- Assume random mating within subpopulations
  - can relax this assumption later.
- Consider the conglomeration of all the subpopulations to be the total population.

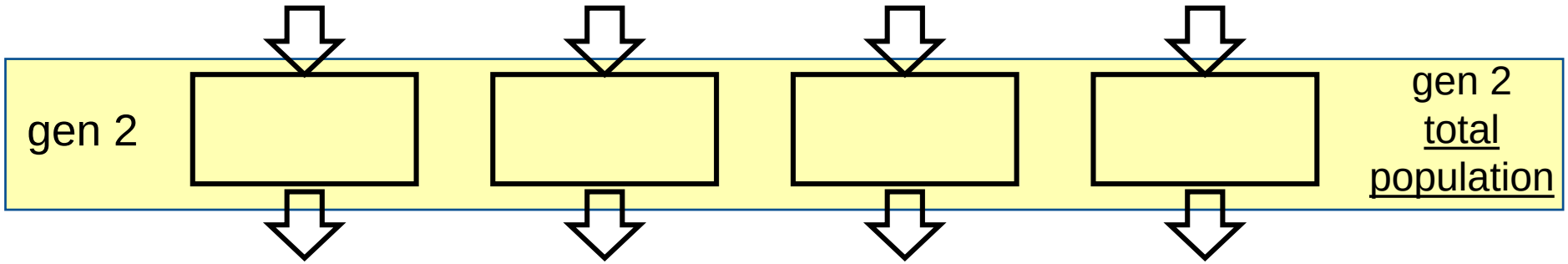
- Consider the conglomeration of all the subpopulations to be the <u>total population</u>.



- Consider the conglomeration of all the subpopulations to be the total population.



- Consider the conglomeration of all the subpopulations to be the total population.
- These individuals don't cross-breed: you simply collect individuals and call the collection the "total population."



(Each one on its own is a subpopulation)

Fragmentation exercise

- Run the inbreeding tool a few (say 5-6) times.
- For each run, pay attention to the amount of genetic variation you see within subpopulations in the first few generations versus the last few generations:
  - how many different alleles there are and the proportion of heterozygous genotypes.

Fragmentation exercise

- For the conglomerate total population, in which generations (first few or final few) does substantial genetic variation exist within subpopulations?

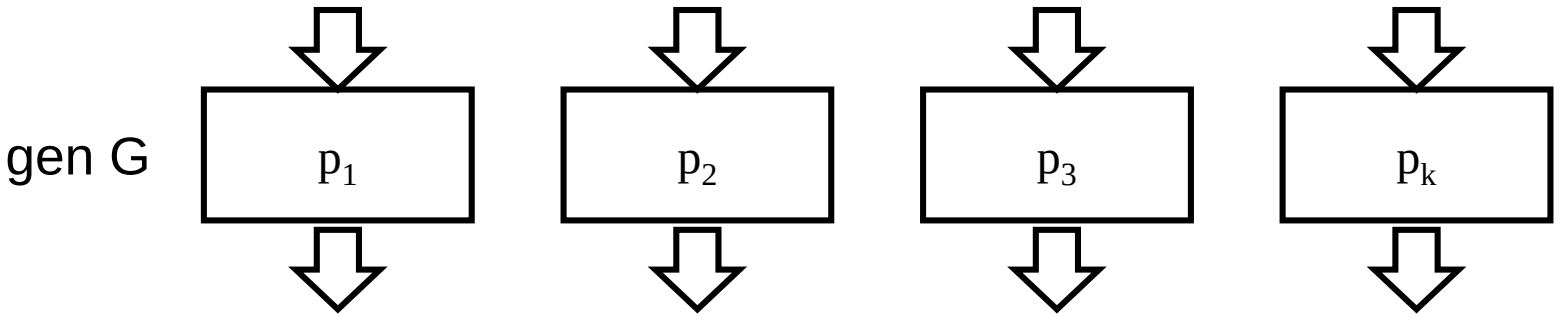
- For which generations does genetic variation exist mainly across the total population (rather than appearing within subpopulations)?

Wright's F statistics

- F_{ST} is the most commonly used.
- Measure of divergence between fragmented subpopulations.
  - Expected to be between 0 and 1.
  - Larger values indicate higher divergence of subpopulations.
- When subpopulations are highly diverged, most of the genetic variation exists at the level of the total population
  - not within subpopulations.

Interpreting F_{ST}

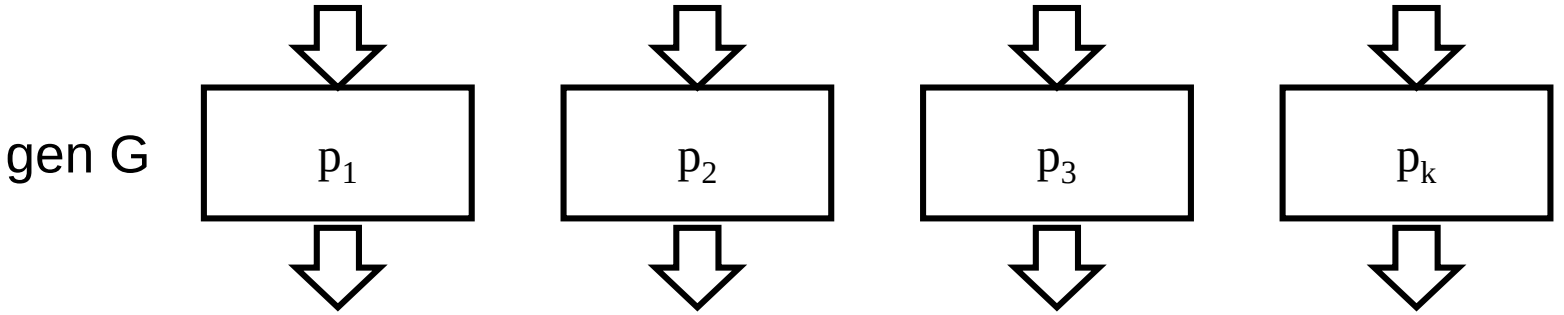
- Measure of variances of allele frequencies within subpopulations …



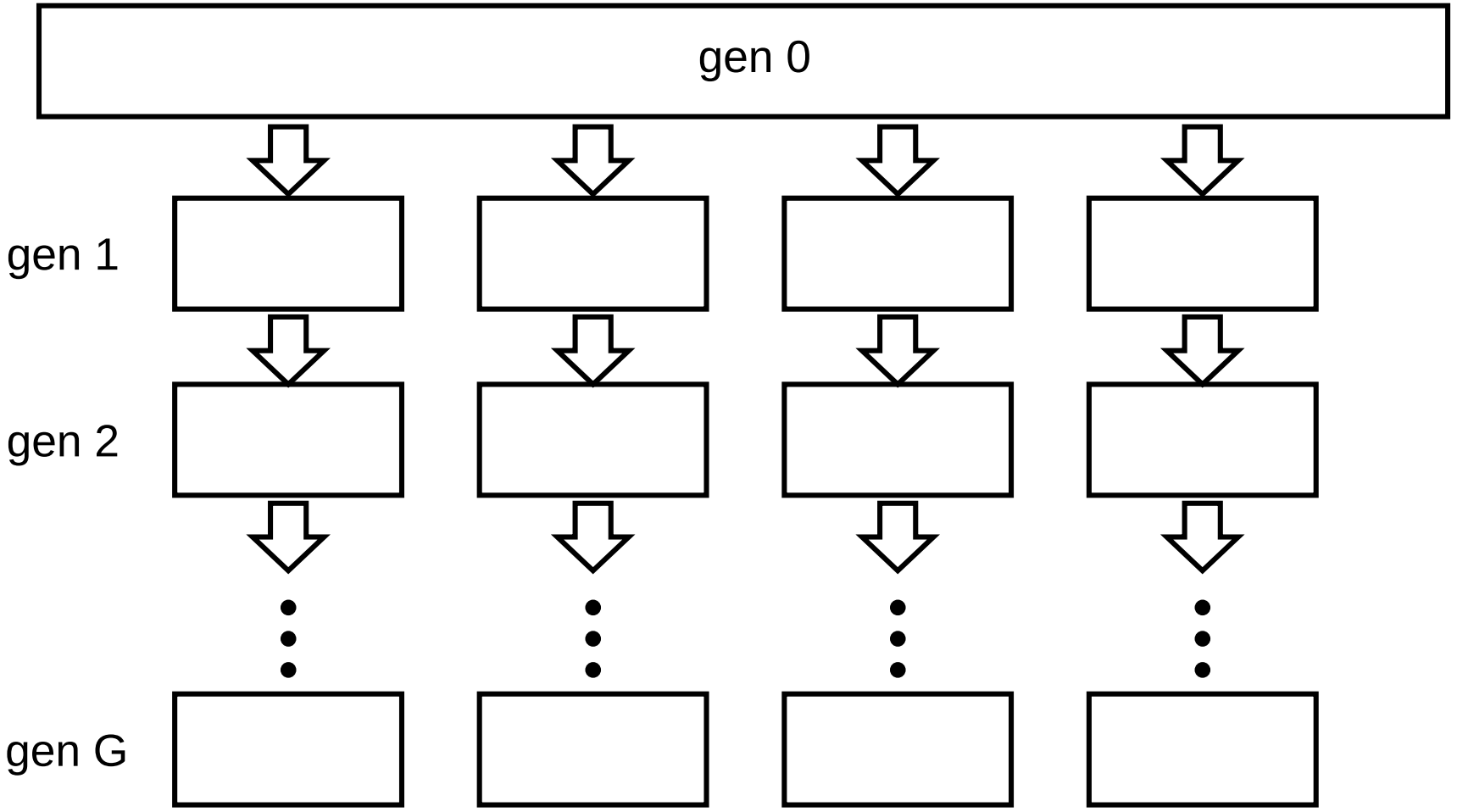
- For k subpopulations, have k allele frequencies (for a given allele), p_1, …, p_k.
- If the allele frequencies are very different between subpopulations, F_{ST} is large.
  - populations have diverged substantially.

Interpreting F_{ST}

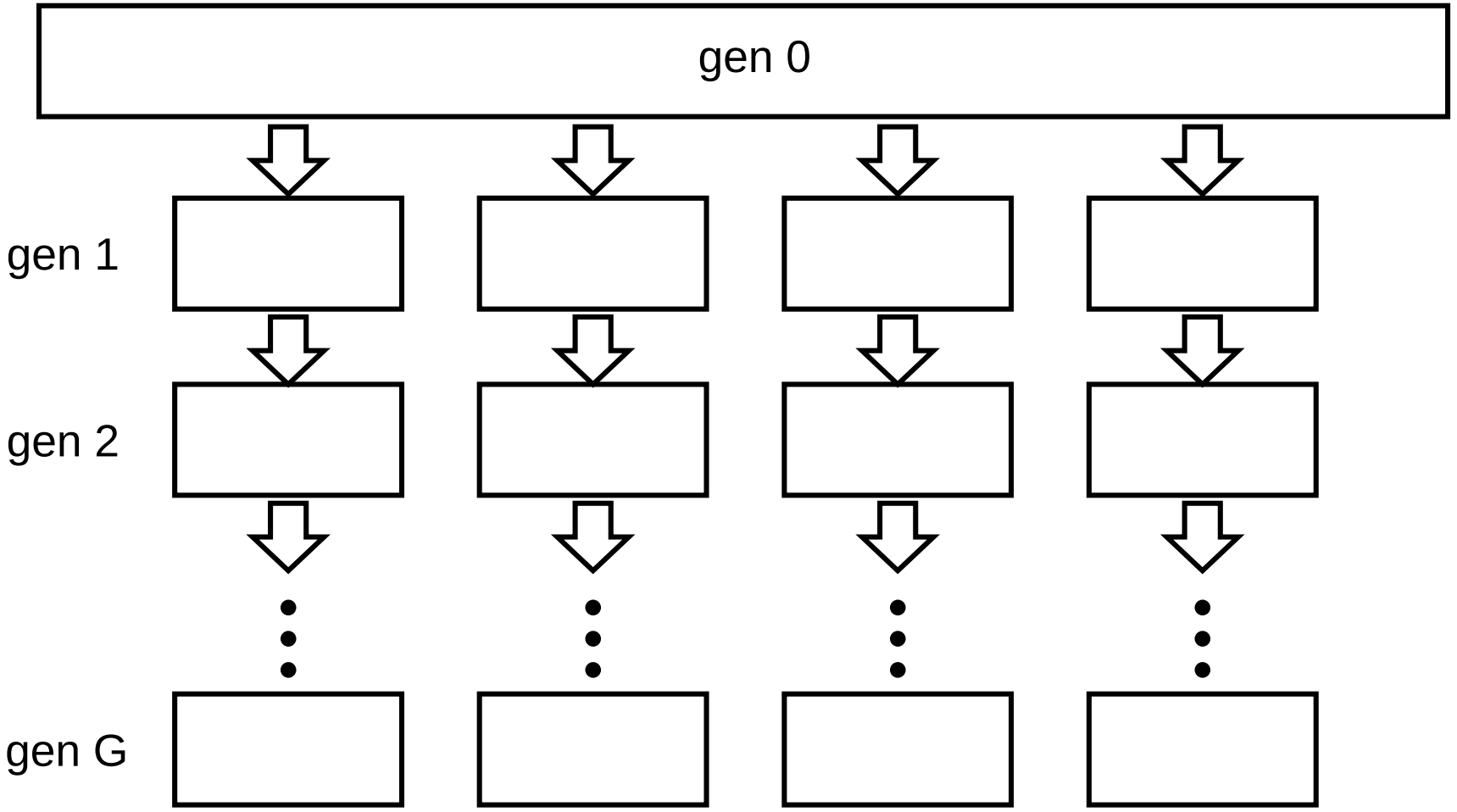
- Measure of variances of allele frequencies within subpopulations …



- For k sub-populations, have k allele frequencies (for a given allele), p_1, …, p_k.
- If the allele frequencies are very similar between subpopulations, F_{ST} is small.
  - populations have not diverged substantially.



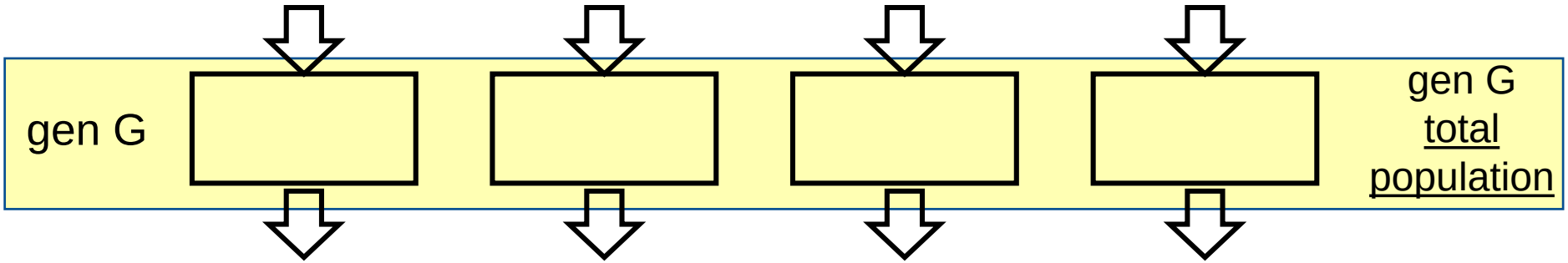
Early in the process (before much time has passed after fragmentation), the subpopulations' genetic composition will be similar to each other. Allele frequencies will be similar (variance will be low).



Later in the process, genetic variation will start to be lost within the individual subpopulations. Which alleles become rarer in each subpopulation is random. Variation in p gets larger.

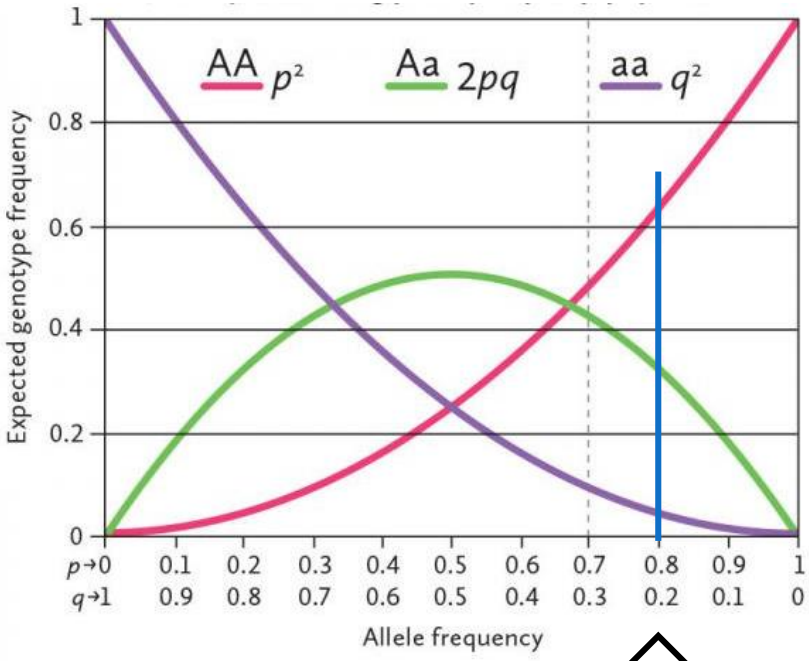
Another way to interpret F_{ST}

- If there is random mating (or close to it) in the subpopulations, we expect to find H-W genotype frequencies within the subpopulations.



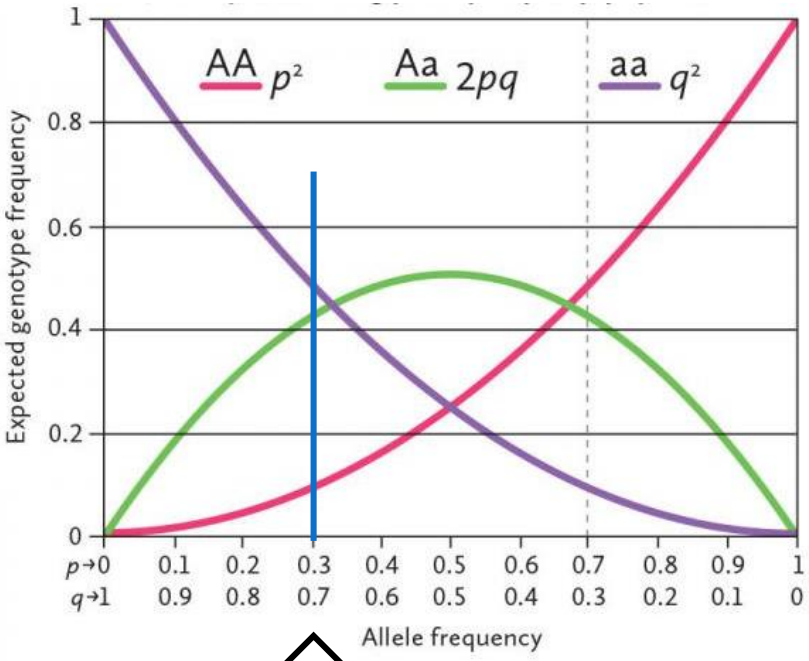
- But, alleles are being fixed and lost within these subpopulations, so allele frequencies are going to their extremes.

H-W genotype frequencies



p in one sub-pop might be here …

A is common in this sub-pop, so **AA** is also common.



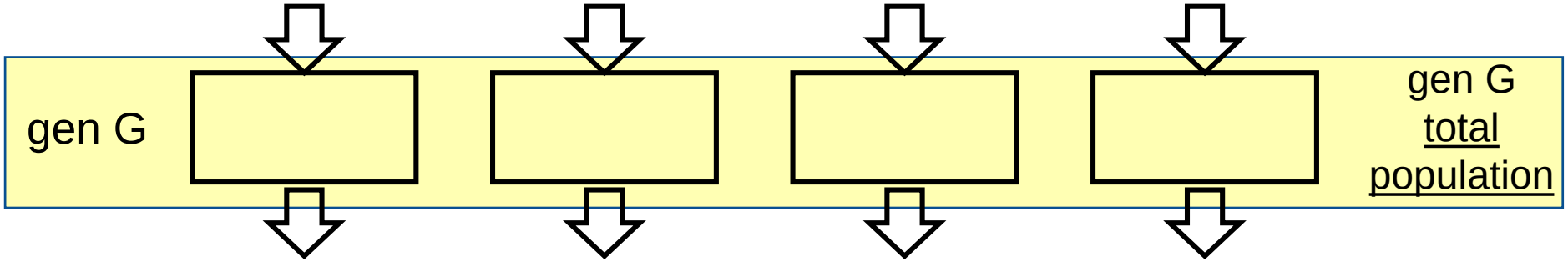
… and in another one p might be here.

A is rarer in this sub-pop, so **AA** is also rarer.

(p was highly similar in both sub-populations at the time of fragmentation)

F_{ST}

- What does the heterozygosity/ homozygosity of the <u>total</u> population look like?



Heterozygosity in the total pop

- Consider a locus with two alleles, A and a.
  - Let's assume a copy of A and a copy of a cannot be IBD.
- So, heterozygous genotypes cannot contain alleles that are IBD.
- What's the frequency of the Aa genotype in the total population?

Heterozygosity in the total pop

- P_{Aa} = Pr (chose two alleles from the population that are not IBD) x Pr (one of them is an A allele) x Pr (one of them is an a allele)

- $P_{Aa} = (1 - F)\ 2p_{tot}q_{tot}$

  F = Fixation index = Pr (two randomly chosen alleles from a population are IBD)

Homozygous frequencies

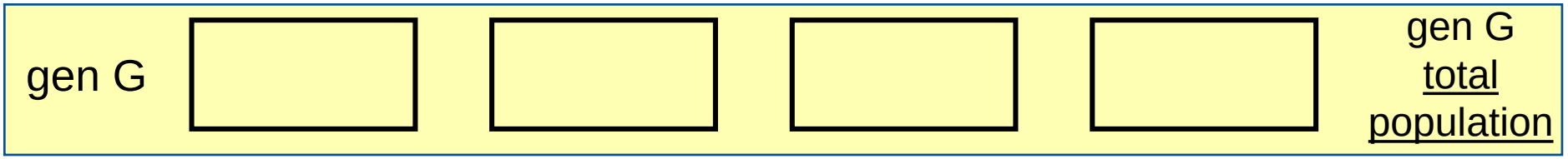
- P_{AA} = Pr (chose two alleles not IBD) x
  Pr (both of them are A) +
  Pr (chose two alleles that are IBD)
  Pr (they are an A allele)

- $P_{AA} = (1 - F)\, p^2_{tot} + F\, p_{tot}$

- $P_{AA} = p^2_{tot} + F\, p_{tot} q_{tot}$

In the total population

- $P_{AA} = p^2_{tot} + F\ p_{tot}q_{tot}$
- $P_{Aa} = 2p_{tot}q_{tot} - F\ 2p_{tot}q_{tot}$
- $P_{aa} = q^2_{tot} + F\ p_{tot}q_{tot}$



In the total population

- $P_{AA} = p^2_{tot} + F\ p_{tot}q_{tot}$
- $P_{Aa} = 2p_{tot}q_{tot} - F\ 2p_{tot}q_{tot}$
- $P_{aa} = q^2_{tot} + F\ p_{tot}q_{tot}$

H-W genotype frequencies

In the total population

- $P_{AA} = p^2_{tot}$ $+ F\ p_{tot}q_{tot}$
- $P_{Aa} = 2p_{tot}q_{tot}$ $- F\ 2p_{tot}q_{tot}$
- $P_{aa} = q^2_{tot}$ $+ F\ p_{tot}q_{tot}$

degree of departure from H-W genotype frequencies

Interpreting F_{ST}

- When considering a fragmented population, the Fixation index for the <u>total</u> population *is* Wright's F_{ST}
- Using the heterozygosity of the total population:

$$P_{Aa} = 2pq - F_{ST}\,2pq$$

$$F_{ST} = \frac{2pq - P_{Aa}}{2pq}$$

(all frequencies are for the total population, this just makes it easier to read)

Interpreting F_{ST}

- $F_{ST} = \dfrac{2pq - P_{Aa}}{2pq}$

(all frequencies are for the total population)

- 2pq is the heterozygosity expected if there is random mating across the total population (no fragmentation).
- P_{Aa} is the actual heterozygosity of the total population.

Wright's F statistics

- Subscripts indicate the level of the population structure from which alleles are being drawn.
- F_{ST} (the most commonly used)
  - "ST" – IBD of alleles w/in Subpopulations with respect to the Total population.
- F_{IT}
  - "IT" – alleles w/in Individuals wrt the Total population.
- F_{IS}
  - "IS" – alleles w/in Individuals wrt the Subpopulation.

F_{IT}

- This is an inbreeding coefficient.
- F_{IT} = Pr (two alleles within an individual are IBD wrt the total population)
  - Sample an individual from the total population, examine the two alleles that individual carries at a locus.
  - Relevant allele frequencies are p_{tot}, q_{tot}.
- If there is random mating within subpopulations, $F_{IT} = F_{ST}$.
  - (under random mating it is equivalent to sample two alleles from a subpopulation or two alleles within an individual in a subpop)

F_{IS}

- This is an inbreeding coefficient.
- F_{IS} = Pr (two alleles within an individual are IBD wrt the subpopulation)
  - Sample an individual from one subpopulation, examine the two alleles that individual carries at a locus.
  - Relevant allele freqs are $p_{subpop(i)}$, $q_{subpop(i)}$.
- If there is random mating within subpopulations, $F_{IS} = 0$.
  - IBD is only considered wrt the previous generation, not further back in time.

Other measures

- G_{ST}
  - Extension of Wrights's F Statistics theory to multiple alleles
    - e.g. microsatellites

- Φ_{ST}
  - Equation structurally similar to F_{ST}, but using nucleotide diversity measures in place of heterozygosity measures.

Hawaiian Petrel

Heredity (Edinb). 2012 Jul; 109(1): 19–28.
Published online 2012 Mar 21. doi: 10.1038/hdy.2012.7

Table 2

Population differentiation of historic and modern Hawaiian petrels based on mitochondrial and nuclear intron data sets

	Hawaii	*Maui*	*Lanai*	*Molokai*	*Kauai*
Hawaii	—	0.092[a]	0.060	NA	0.064[a]
Maui	0.068[a]	—	0.095[a]	NA	−0.030
Lanai	0.405[a]	0.543[a]	—	NA	0.145[a]
Molokai	0.226[a]	0.404[a]	0.037	—	NA
Kauai	0.511[a]	0.574[a]	0.633[a]	0.424[a]	—

Abbreviation: NA, not available.

Pairwise F_{ST} values for the *Cytochrome b* gene are below the diagonal, whereas those for a data set of sequences from three nuclear introns are above.

[a]Indicates the estimate is significantly different from zero after correction for multiple tests.

The Condor 99:812–815

GENETIC DIVERSITY AND DIVERGENCE OF ENDANGERED GALÁPAGOS AND HAWAIIAN PETREL POPULATIONS[1]

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- Based on a small number of markers, the authors estimated F_{ST} between Galapagos and Hawaiian petrels to be 1. Their conclusion is that these should be treated as different species of petrels.

Sierra Nevada Red Fox

- Native range being infiltrated by exotic populations.

Sierra Nevada Red Fox

Perrine, et al.

Conserv Genet (2007) 8:1083–1095

Table 3 Pairwise F_{ST} and Φ_{ST} estimates among three historic (pre-1950) and four modern (post-1950) California red fox populations

	Historic SN	Historic CS	Historic SV	Modern CS	Modern SV	Modern BA	Modern SC
Historic SN[a]	–	–0.08	0.51*	0.00	0.45*	0.36*	0.27*
Historic CS	–0.10	–	0.51	0.22	0.41*	0.18	0.11
Historic SV	0.42*	0.42	–	0.75*	–0.06	0.40*	0.32*
Modern CS	0.06	0.22	0.73*	–	0.54*	0.31*	0.21
Modern SV	0.54*	0.60*	–0.06	0.80*	–	0.40*	0.33*
Modern BA	0.44*	0.39*	0.40*	0.65*	0.51*	–	0.09
Modern SC	0.38*	0.31*	0.33*	0.56*	0.44*	0.18	–

Below diagonal measures are based solely on haplotype frequencies (F_{ST}); above diagonal estimates incorporate pairwise differences in sequence divergence (Φ_{ST})

[a] SN = Sierra Nevada, CS = Cascades, SV = Sacramento Valley, BA = San Francisco Bay Area, SC = Southern California

* significant at $\alpha = 0.05$ using sequential Bonferroni correction for multiple tests (Rice 1989)

- Native and exotic populations appear to remain distinct, and native populations are not highly diverged from one another.

Genetic rescue

- Populations with historically large habitat ranges now existing in smaller fragmented populations may benefit from crossing between populations.
  - Artificially manage gene flow between subpopulations.
  - Has the potential to counter inbreeding depression within populations.

Rocky Mountain Bighorn Sheep

Rocky Mountain Bighorn Sheep

- Population established on the National Bison Range in Montana in 1922
  - isolated until 1985
- In 1985, animals derived from two outbred herds were introduced.
- Analysis of data collected from 1979-2003 (Hogg, et al. 2006, PNAS 273:1491-1499)
  - Reported a net positive effect of outbreeding in both males and females for a number of traits observed, including major components of fitness.

Hogg, et al. 2006,
PNAS 273:1491-1499

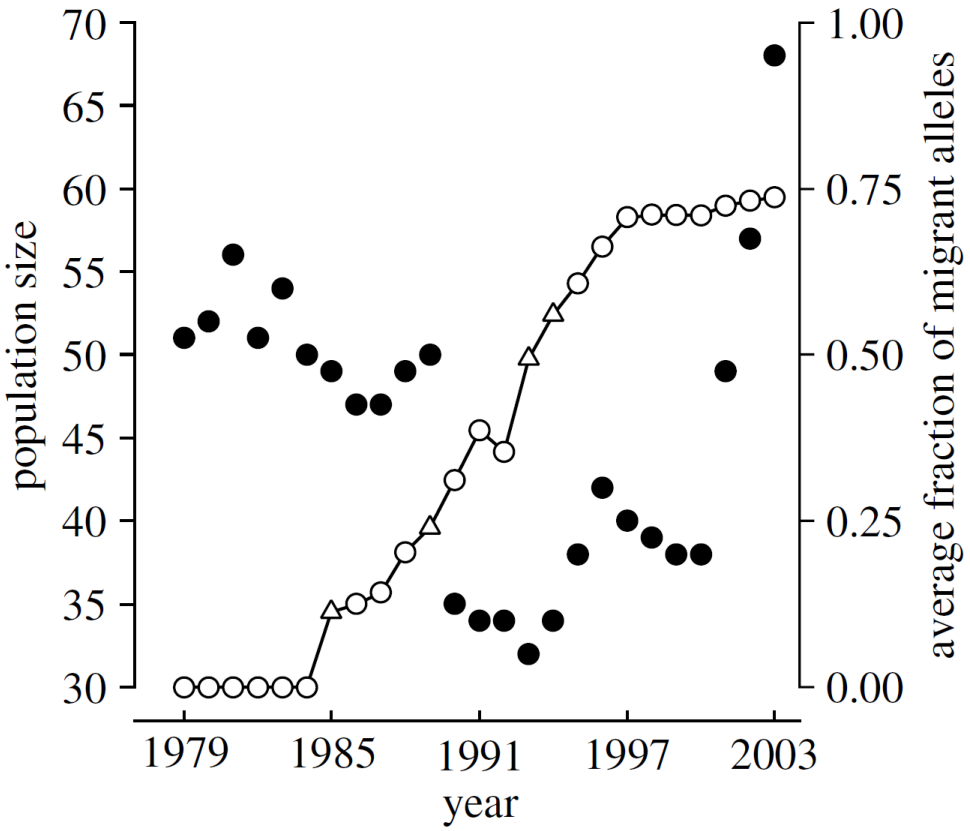

Figure 3. Total number of native bighorn (residents minus migrants; filled symbols) in relation to the average fraction of migrant alleles carried by resident bighorn (natives plus migrants; open symbols) during 1979–2003. Reversal of a multi-year decline in population size coincided with the year (1993) in which the average resident carried a majority of migrant alleles. Because migrants were excluded from annual population number (filled symbols, left axis), population changes are entirely due to reproduction and mortality within the NBR population. Years in which migrants arrived are

Outbreeding depression

- If populations are highly adapted to their local environment, bringing in non-beneficial genetic variants may reduce fitness of the population.
- If populations are substantially diverged, hybrid incompatibility may occur.
- Small number of published examples.

Arabian Oryx

Arabian Oryx

- Original habitat extended across the Arabian peninsula.
- Hunting led to severe decline of natural population.
- Extinction in the wild in 1972.
- Captive breeding in zoos in the 1960s
  - animals had been collected from various diverse locations.
- Reintroduction of animals to the wild beginning in the 1980s.

Arabian Oryx

- Analysis of juvenile mortality was performed over the next few years
  - Marshall & Spalton, Animal Conservation (2000) 3:241–248.
- Conclusions were that the reintroduced population suffered both inbreeding and outbreeding depression
  - Juvenile mortality was associated with both high levels of inbreeding and with high levels of heterozygosity.

Genetic rescue vs outbreeding depression

- Published examples of inbreeding depression are numerous.
- Many experiments have demonstrated the ability to reverse inbreeding depression.
  - Only a handful of published cases of genetic rescue for conservation purposes exist.
- Few examples of outbreeding depression in natural populations have been published.
- Methods to predict outbreeding depression have been developed (e.g. Frankham, Molec. Ecol. 2015)
  - Expectation is that natural selection will ultimately overcome outbreeding depression.