Chapter 6

Population Genetics and Evolution: A Simulation Exercise

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Introduction

Basic population genetics concepts are often poorly understood by introductory biology students. In some cases, the student finds it difficult to recognize that the Hardy-Weinberg law describes an ideal population and provides biologists with a standard against which evolutionary changes can be measured. In other situations, students have problems visualizing the types of changes that can occur in the population gene pool when microevolutionary forces, such as mutations, genetic drift, gene flow, and natural selection, are in effect. For introductory biology students, the types of changes associated with these microevolutionary forces are not readily observed through experimental exercises. Simulation activities provide the best means for reinforcing these topics at the introductory level.

This lab exercise, which was developed several years ago for the students enrolled in our introductory biology course, involves both a hands-on and a computer simulation activity. The overall objectives of the lab are 1) to become familiar with the premises of the Hardy-Weinberg Law, 2) to understand the types of forces that cause microevolutionary changes in natural populations, and 3) to practice using the Hardy-Weinberg principle to demonstrate evolutionary change.

A particularly appealing feature of the hands-on simulation is that students work as part of a team, with four different teams acting as "researchers" on an island ecosystem. Throughout the course of the lab exercise, students are expected to engage in significant discussions both within a single team and among the four island teams. The students enjoy the simulation activities and they feel that this lab effectively improves their understanding of population genetics. The laboratory exercise works well in a lab of 12-24 students. Both the hands-on simulation and the computer exercise can easily be completed within a three hour lab period; the hands-on simulation alone could be finished in a 2 hour lab. The set-up time is minimal (30-60 minutes) and the materials are completely reusable.

Materials

Each "island" will need the following supplies (12-16 students per island). Bead suppliers are listed in the appendix.

10,000 4 mm faceted plastic beads, pink color
10,000 4 mm faceted plastic beads, white color
10,000 4 mm faceted plastic beads, red color
10,000 4 mm faceted plastic beads, green color
400 11 mm plastic tri beads (triangular shape), pink color
400 11 mm plastic tri beads (triangular shape), white color
400 11 mm plastic tri beads (triangular shape), red color
4 plastic shoe boxes (13" x 7" x 4")
4 0.25 inch mesh sieves (Construction details are in the appendix.)
4 pairs of forceps
4 stopwatches

Notes for the Instructor

In our introductory lab, a single lab bench is used to represent each island. We store the faceted beads in the shoe boxes and label the top according to the geographical location associated with the color of the beads. At each island, place all of the faceted beads of a single color into one of the four shoe boxes. Label the box with the pink beads "EAST", the box with the red beads " NORTH", the box with the white beads "SOUTH", and the box with the green beads "WEST". Place the four boxes with beads along the four sides of your designated "island". Place the tri beads into three small zip lock bags—you should put about 100 beads of a single color into each bag. At least 2 (and as many as four) students should be assigned to each shoebox station. With labs of more than 16 students, more than one island will need to be set up. At each "shoebox" station, you should put the following materials:

one Ziplock™ bag with 100 red tri beads
one Ziplock™ bag with 100 pink tri beads
one Ziplock™ bag with 100 white tri beads
one pair of forceps
one stopwatch
one sieve

To make the students feel like they are working on a tropical island, we decorate the lab. We have constructed mountains to separate the geographical regions of the islands. Since there are fairly tall mountains that separate the southeastern part of the island from the northwestern region, the students can readily visualize that gene flow is most likely to occur EAST → SOUTH or WEST → NORTH. We also use poster board to create beaches that have colored sand that matches the color of the faceted beads at each geographical location.

Student Outline

Theory

Population genetics is a branch of biology concerned with genetic changes in populations--in particular, changes in a gene pool over time. You should recall that a gene pool is the collection of all genes carried by the individuals in the population. At any particular time, the gene pool of a population is described in terms of both gene and genotype frequencies.

Just as Mendel's laws form the cornerstone for classical genetic studies, the Hardy-Weinberg principle forms the cornerstone for population genetic studies. To appreciate fully the significance of the Hardy-Weinberg principle, we must first understand how evolutionary agents operate. You should recall the major evolutionary forces that can alter population gene pools include:

1. mutations
2. emigration/immigration (gene flow)
3. random genetic drift
4. natural selection

Let us now consider each of these evolutionary agents as it applies to population genetics. *Mutations* refer to biochemical changes in the genetic material of an organism. Mutation rates commonly occur in natural populations at a fairly low rate, yet they are ultimately the most important source of genetic variation in the population gene pool. *Gene flow* refers to the emigration or immigration of genes out of or into the population. *Random genetic drift* occurs when unpredictable variations change the structure of the gene pool. The effects of random genetic drift are minimized as the size of the breeding population increases. *Natural selection* occurs when a particular phenotype has a greater (or lesser) reproductive success than an alternate phenotype. Reproductive success is measured by an individual's ability (a) to survive to reproductive age and (b) to produce viable offspring. The effects associated with natural selection can also occur if random mating is not prevalent. Random mating requires that the probability of any two alleles coming together in the fertilized egg is only related to the probability of randomly encountering each allele in the gene pool.

Now that we understand the types of evolutionary forces that can operate in a population, we can turn our attention to the specifics of the Hardy-Weinberg principle. Under the conditions of the Hardy-Weinberg principle, we assume that the gene pool is a closed entity that is not affected by any of the evolutionary agents discussed above. Given this assumption, the Hardy-Weinberg principle states that there will be no change in either the gene or genotype frequency of the population from one generation to the next. Clearly, the conditions for a Hardy-Weinberg equilibrium are idealized. In most human populations, emigration and immigration are a way of life and random mating is certainly uncommon. We tend to choose as mates people with similar cultural (and to some degree similar genetic) backgrounds. The probability that any of you will mate with an Australian aborigine is nearly zero. So, why is the Hardy-Weinberg principle so important? The answer: When the conditions for Hardy-Weinberg equilibrium are *NOT* met, the resulting *difference* between the actual gene frequencies and those expected under ideal Hardy-Weinberg conditions allows us to determine the *rate* of evolutionary change.

Let us now consider a single genetic locus with two alleles, A and a. Let

p = frequency of occurrence of the A allele
q = frequency of occurrence of the a allele

Given the p and q allele frequencies, the Hardy-Weinberg Principle allows us to calculate the genotype frequencies in the population as follows:

p^2 = frequency of occurrence of the AA genotype
2pq = frequency of occurrence of the Aa genotype
q^2 = frequency of occurrence of the aa genotype

Derivation of the Hardy-Weinberg Formulas

Although you may not realize it, the genotype frequencies calculated for a population that is in Hardy-Weinberg equilibrium can be derived using a simple Punnett Square. Let's go through this derivation to give you a better understanding of the mathematical basis underlying the Hardy-Weinberg Law.

GIVEN**:** Let us assume that we are studying a population of the peppered moth, *Biston betularia*, living in the English countryside. As you may recall, the dark form of this moth carries a dominant allele ("A") while the peppered form of the moth carries two recessive alleles ("a"). In polluted regions of England, the dark form of the moth is most common since the pollution effectively eliminates the lichen that commonly grows on the tree trunks. Under these conditions, the dark moth is most likely to survive and reproduce since it is not readily eaten by the bird predators while it rests on the trunks of the trees. In unpolluted regions, where the tree trunks are covered with lichen, the peppered moths are most successful since they blend in very well against the lichen-covered tree trunks. In the population that we are studying, the frequency of the dark allele ("A") is .30 and the frequency of the peppered allele ("a") is .70.

ASSUMPTIONS: The population under consideration is in Hardy-Weinberg equilibrium and the "A" and "a" alleles are not carried on the sex chromosomes.

Under these conditions, any offspring in the population has a .30 chance of receiving the dark ("A") allele from either parent and a .70 chance of receiving the peppered ("a") allele. Recall that we are dealing with an entire population and these values represent average probabilities across the entire population. We can calculate the expected genotype frequencies among all of the offspring in this population:

		MALE	GAMETE
		"A" ALLELE (Frequency = .30)	**"a" ALLELE (Frequency = .70)**
Female	**"A" ALLELE (Frequency = .30)**	AA (.30)(.30) = .09	Aa (.70)(.30) = .21
Gamete	**"a" ALLELE (Frequency = .70)**	Aa (.70)(.30) = .21	aa (.70)(.70) = .49

The overall probability for each expected genotype in this population is:

homozygous AA (dark moths) = .09 (p^2)
heterozygous Aa (dark moths) = .21 + .21 = .42 (2pq)
homozygous aa (peppered moths) = .49 (q^2)

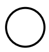
If a population is in Hardy-Weinberg equilibrium, you can calculate the expected genotypic frequencies given the allele frequencies AND all of these frequencies will remain constant from one generation to the next as long as equilibrium conditions persist. Thus, the Hardy-Weinberg principle simply demonstrates that 1) both dominant and recessive alleles can be maintained in a population and that 2) recombination occurring as a result of meiosis and fertilization does not by itself change the overall composition of the gene pool.

Procedure

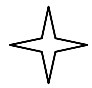
The Gene Pool

In this exercise, you will be working with a population of GRAVEL DIVERS. Gravel divers, which look very much like plastic tri beads, can exist in one of three different colors. The red gravel divers (AA) are homozygous for the "A" allele; the white (clear) gravel divers (aa) are homozygous for the "a" allele; the pink gravel divers (Aa) are heterozygous for the two alleles. Gravel divers normally bury themselves in gravel, which looks very much like plastic faceted beads. As far as we know, these gravel divers are only found on three islands, each one of which looks very much like a lab bench. On the exotic islands where these gravel divers are found, the color of the gravel varies according to geographical location. Specifically, on the north side of the island, the gravel is red; on the east side of the island, the gravel is pink; on the south side of the island, the gravel is white (clear), and on the west side of the island, the gravel is green. The native predator of the gravel diver is an organism called the GRAVEL DIVER PICKER. This organism, which looks very much like you and your lab partners, looks for gravel divers that are buried in the gravel and picks them out of the gravel with a pair of forceps.

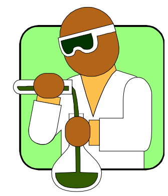
Now, let's review the essential information that you have been given concerning the study organism and its surrounding environment. The following illustrations should help you remember who is who:



GRAVEL



GRAVEL DIVER



GRAVEL DIVER PICKER

The geographical differences in the gravel on the island will become important. The following diagram will help you identify the major geographical locations on the island:

NORTH *(red gravel)*

WEST *(green gravel)*

EAST *(pink gravel)*

SOUTH *(white gravel)*

Now, let's proceed to study the gene pool of the gravel diver population and the impact of various evolutionary forces on this gene pool.

Creating the equilibrium population

- You have been given a plastic box containing gravel that is representative of the specific geographical region of the island that you are studying. This plastic box represents your study site. Remember that there are four different study sites (N, S, E, W) being researched on each of three different islands in the two lab rooms. Look at the color of your gravel (round beads) and determine the location of your study site (North, South, East or West). Find the other lab groups working at the three other sites (as determined by different colors of gravel) on your island. Throughout the lab, you will be working closely with the other researchers on your particular island.

- Into your gravel, add a total of 100 gravel divers. Use the following table to determine the number of gravel divers of each color that you should add to your gravel. The initial frequency of the red, pink, and white gravel divers varies according to the specific geographical location on the island.

Number of Gravel Divers of Each Color to be added to Geographical Gravel Location

	RED	*PINK*	*WHITE*
NORTH *(red gravel)*	64	32	4
EAST *(pink gravel)*	49	42	9
SOUTH *(white gravel)*	36	48	16
WEST *(green gravel)*	25	50	25

- Gravel divers like to burrow in the gravel, so be sure to bury your gravel divers in the gravel thoroughly. Since the color of the gravel divers is an example of co-dominance, the genotype and the phenotype frequencies are the same. Calculate the *genotype and phenotype frequencies* for the gravel divers in your population using the following formula:

$$\text{Frequency of Red (AA) Gravel Divers} = \frac{\text{Number of Red gravel divers}}{\text{Total Number of gravel divers}}$$

- Repeat this calculation to determine the genotype/phenotype frequencies of the pink and white gravel divers in your population. Fill in these initial genotype/phenotype frequencies in Table 6.1 below.

- Calculate the *allele frequencies* for the gravel diver population . The alleles in our population are codominant. That is, each red gravel diver carries two red alleles ("A"); each white gravel diver carries two white alleles ("a"); each pink gravel digger carries one red ("A") and one white ("a") allele. To calculate the frequency of the red ("A") allele in the gravel diver population, perform the following calculation:

$$p = \frac{(2)(AA) + Aa}{(2)(N)}$$

where, p = red allele frequency in the gravel diver population
AA = TOTAL NUMBER of red gravel divers in your population
Aa = TOTAL NUMBER of pink gravel divers in your population
N = TOTAL NUMBER of ALL gravel divers in your population

Since you only have two different alleles at the genetic locus under consideration, you can easily calculate the frequency of the white ("a") alleles as follows:

$$q = 1.0 - p$$

where, q = white allele frequency in the gravel diver population

Table 6.1. Initial frequencies for your gravel diver population**.**

YOUR STUDY SITE:______________________ (Geographical Location)

GENOTYPE/PHENOTYPE FREQUENCIES | *ALLELE FREQUENCIES*

RED (AA)	PINK (Aa)	WHITE (aa)	RED (p) (“A” allele)	WHITE (q) (“a” allele)

- If we now leave these gravel divers alone, they will (hopefully) survive and multiply. You should recognize that our gene pool is a closed entity and the probability of two gravel divers mating is only related to their frequency of occurrence in the population. Presently, there are no external forces (such as immigration/emigration, natural selection, mutation, etc) operating on our population. As such, our population, ideal as it is, conforms to the assumptions of the Hardy-Weinberg principle. That means that we can use the following equations to calculate the genotype frequencies in our population:

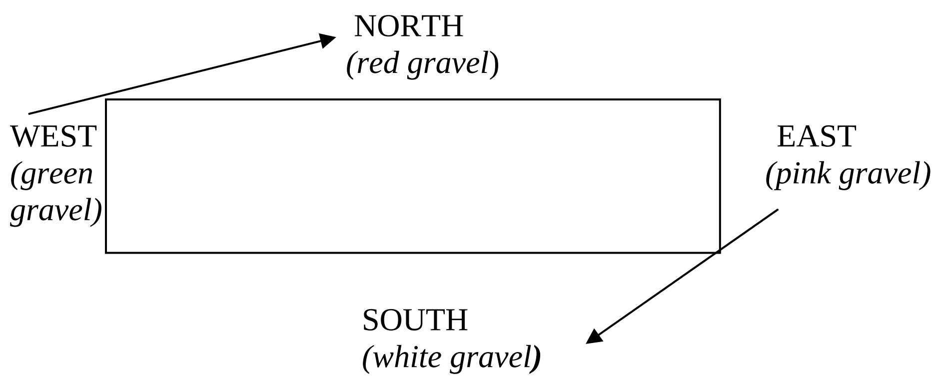
p^2 = frequency of red gravel divers
$2pq$ = frequency of pink gravel divers
q^2 = frequency of white gravel divers

- Use the “p” and “q” values that you entered in Table 1 above to calculate the EXPECTED genotype frequencies given the assumptions of the Hardy-Weinberg principle. Compare your EXPECTED frequencies to the OBSERVED frequencies that you entered in Table 6.1. Is your population in Hardy-Weinberg equilibrium? Why or Why not?

Gene Flow

Spring has come to your island and the reproductively mature gravel divers are anxious to find a mate. The gravel divers at the four geographical locations on your island tend to behave somewhat differently during mating season. Some biologists think that these differences are significant enough to suggest that these populations may actually be subspecies (but we won’t get into gravel diver classification in this lab). The gravel divers living on the North and the South sides of the island are basically happy homebodies. They like the other gravel divers in their own population and they just find a neighborhood male or female with which they are compatible and mate. The gravel divers living on the East and the West sides of the island are more adventurous. Many of these gravel divers commonly leave their natal population to find a mate.

Previous studies have shown that due to the presence of mountain ranges within the island, some of the gravel divers from the eastern shore will migrate to the southern shore and some of the gravel divers from the western shore will migrate to the northern shore. In case you are thoroughly confused, the following diagram will help you visualize the springtime migration patterns that occur on your island:



This annual migration results in gene flow among the populations on the island. Gravel divers are emigrating away from the eastern and western sites and immigrating into the northern and southern sites respectively. We are now violating one of the basic assumptions of the Hardy-Weinberg principle which means that a microevolutionary agent is now operating on this island.

You have joined a research team that is trying to determine the impact of this gene flow (emigration or immigration) on your original population.

❖ *Generating a Hypothesis*: "What do you expect to happen at YOUR study site?"
For each of the following genotype and allele frequencies, clearly indicate whether you expect the frequency to increase (+), decrease (-), or remain the same (0) AFTER migration has occurred AT YOUR STUDY SITE.

EXPECTED FREQUENCY CHANGES

		GENOTYPE		*ALLELE*	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (A)	WHITE (a)
Expected Changes (+, -, 0)	_______	_______	_________	_______	________

❖ If your original study population is on the EAST side (i.e. you have PINK gravel), some of your gravel divers will migrate to the south. Use the sieve to simulate emigration. Sift the gravel with your sieve one time. Remove the gravel divers that are sifted out with the sieve, count the total number of gravel divers of EACH COLOR that you remove. Give these emigrants to one of the researchers associated with the southern population (WHITE gravel) on your island.

❖ If your original study population is on the WEST side (i.e. you have GREEN gravel), some of your gravel divers will migrate to the north. Use the sieve to simulate emigration. Sift the gravel with your sieve one time. Remove the gravel divers that are sifted out with the sieve, count the total number of gravel divers of EACH COLOR that you remove. Give these emigrants to one of the researchers associated with the northern population (RED gravel) on your island.

❖ At this point, all of you should have either lost or gained some gravel divers through emigration or immigration. Fill in Table 6.2 to reflect these changes in your gene pool:

Table 6.2. The number of gravel divers of each color before and after gene flow.

	NUMBER OF GRAVEL DIVERS OF EACH COLOR			
	RED (AA)	*PINK(Aa)*	*WHITE(aa)*	*TOTAL*
Before gene flow				
Number added (immigrants) or removed (emigrants)				
After gene flow				

❖ Calculate your new allele and genotype frequencies as follows:

Frequency of Red ("A") allele:

$$p = \frac{(2)(AA) + Aa}{(2)(N)}$$

Frequency of White ("a") allele:

$$q = 1.0 - p$$

Genotype frequencies:

Red gravel divers ("AA") = AA/N
Pink gravel divers ("Aa") = Aa/N
White gravel divers ("aa") = aa/N

where AA = number of red gravel divers remaining after gene flow,
Aa = number of pink gravel divers remaining after gene flow,
aa = number of white gravel divers remaining after gene flow, and
N = total number of gravel divers remaining after gene flow.

- Fill out Table 6.3 to calculate the effect that gene flow had on the allele and genotype frequencies in your population.

Table 6.3. *Genotype/phenotype and allele frequencies for your gravel diver population before and after gene flow.*

YOUR STUDY SITE:______________________ (Geographical Location)

	GENOTYPE FREQUENCIES			ALLELE FREQUENCIES	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (p) (“A” allele)	WHITE(q) (“A” allele)
Initial Population					
After Gene Flow					

- *Drawing a Conclusion*. Compare the observed results recorded in the above table with the expected results recorded in the Expected Frequency Changes table. Explain any discrepancies that you saw between the recorded observations and the expected results.

- Is your population still in Hardy-Weinberg equilibrium? Use the “p” and “q” allele frequencies that you calculated AFTER gene flow (in Table 6.3 above) to calculate the genotype frequencies that you would EXPECT to observe under the assumptions of the Hardy-Weinberg principle. Remember, that means that we can use the following equations to calculate the genotype frequencies in our population:

p^2 = frequency of red gravel divers,
$2pq$ = frequency of pink gravel divers, and
q^2 = frequency of white gravel divers.

- Compare these EXPECTED genotype frequencies to the OBSERVED genotype frequencies from Table 6.3. Are they identical? Provide a biological explanation for these results.

- After you have finished simulating the effects of gene flow, get together with the groups studying the gravel diver populations in the other three geographical regions of your island and discuss the following questions:
    a) What effect did the gene flow have on the original allele and genotype frequencies in the gravel diver populations living in the North? East? South? West?
    b) Did your observations in part (a) agree with your expectations? Be sure that you can explain any deviations between observed and expected results.

c) In this simulation, you were told to SELECT a certain color of gravel diver that underwent immigration/emigration. What results would you have expected if a RANDOM sample of your gravel divers immigrated/emigrated? Based on these observations/thoughts, can you draw any conclusions as to the conditions under which gene flow would be an especially strong evolutionary agent? Does gene flow ALWAYS change the gene pool of a population?

Natural Selection

The heat of the mating season has finally subsided and the gravel diver populations are once again putting their efforts into survival and growth. In the absence of unusual environmental changes, the major factor affecting the survival of the gravel divers appears to be predation. The major predator of the gravel divers is the gravel diver picker (known as a GDP for short). As we have previously noted, the GDP looks very much like you and your lab partners. As you might guess, the ability of the GDP to prey successfully upon the gravel divers depends on both the color of the gravel diver and the color of its environment (the gravel). In other words, unless your study site is on the western side of the island (GREEN gravel), some of the gravel divers are more readily camouflaged by the gravel than others. For example, to find a red gravel diver on the north side of the island requires careful scrutiny of the red gravel. In evolutionary terms, we recognize that some colors of gravel divers have a higher fitness and are more likely to escape predation than other colors. These gravel divers are more likely to survive to reproductive age and contribute to the gene pool in the next generation. In addition, the relative fitness associated with a particular phenotype varies with the environment. Over time, we would expect that the frequency of the gravel divers with the greatest fitness will increase IF the environment remains constant. In other words, with a strong predation pressure from the gravel diver pickers, we would expect the frequency of red gravel divers to increase on the north side of the island, pink gravel divers would become most prevalent on the eastern side of the island, and white gravel divers would flourish on the southern end of the island. In this part of the exercise, you will study the change in the gene pool that occurs due to the predatory activities of the GDP on the gravel divers.

- *Generating a Hypothesis*: **"**What do you expect to happen at YOUR site?"

  At my site, __________ (RED, WHITE, PINK, ALL) gravel divers will have the greatest chance of surviving predation by the gravel diver picker.

- For each of the following genotype and allele frequencies, clearly indicate whether you expect the frequency to increase (+), decrease (-), or remain the same (0) AFTER natural selection has occurred AT YOUR STUDY SITE.

EXPECTED FREQUENCY CHANGES

	GENOTYPE			*ALLELE*	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (A)	WHITE (a)
Expected Changes (+, -, 0)	_______	_______	_________	_______	________

- ❖ Choose one person in your lab group to be the predator (GDP). This person MUST use a pair of forceps when preying upon the gravel divers. The use of forceps simulates the time it takes a GDP to find and immobilize a gravel diver. Also, GDP's can only capture one gravel diver at a time.

- ❖ Choose a second person in your lab group to be the timekeeper. GDP's can only prey on the gravel diver population for a total of 1 minute at a time. We will assume that after 1 minute of predation, the GDP needs to stop and eat its prey before the gravel divers that have been caught come to their senses and escape. Thoroughly mix up both the gravel and the gravel divers in your plastic box.

- ❖ When the timekeeper says GO, the GDP should remove gravel divers from the gravel. Remember to remove ONE AT A TIME and you MUST USE FORCEPS to remove each gravel diver. The object is to remove as many gravel divers as you can in 1 minute.

- ❖ After the minute has passed, count the number of gravel divers removed and complete Table 6.4.

Table 6.4. *The number of gravel divers of each color before and after natural selection.*

	NUMBER OF GRAVEL DIVERS OF EACH COLOR *RED (AA)*	*PINK(Aa)*	*WHITE(aa)*	*TOTAL*
Before natural selection				
Number removed by predation				
Number remaining after natural selection				

- ❖ The gravel divers that were eaten by the GDP should be removed permanently from the population that you are studying.

❖ Calculate your new allele and genotype frequencies as follows:

Frequency of Red ("A") allele:

$$p = \frac{(2)(AA) + Aa}{(2)(N)}$$

Frequency of White ("a") allele:

$$q = 1.0 - p$$

Genotype frequencies:

Red gravel divers ("AA") = AA/N,
Pink gravel divers ("Aa") = Aa/N, and
White gravel divers ("aa") = aa/N.

where AA = number of red gravel divers remaining after predation,
Aa = number of pink gravel divers remaining after predation,
aa = number of white gravel divers remaining after predation, and
N = total number of gravel divers remaining after predation.

❖ After you have calculated the new allele and genotype frequencies, fill out Table 6.5 to determine the effect that natural selection in the form of predation had on your population.

Table 6.5. *Genotype/phenotype and allele frequencies for your gravel diver population before and after natural selection.*

YOUR STUDY SITE:**_______________________** (Geographical Location)

	GENOTYPE FREQUENCIES			*ALLELE FREQUENCIES*	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (p) (“A” allele)	WHITE(q) (“A” allele)
Initial Population					
After Gene Flow **(***from Table 3*)					
After Natural Selection					

❖ *Drawing a Conclusion.* Compare the observed results recorded in the above table with the expected results recorded in the Expected Frequency Changes table. Explain any discrepancies that you saw between the recorded observations and the expected results.

Is your population still in Hardy-Weinberg equilibrium? Use the “p” and “q” allele frequencies that you calculated AFTER gene flow (in Table 6.5 above) to calculate the genotype frequencies that you would EXPECT to observe under the assumptions of the Hardy-Weinberg principle. Remember, that means that we can use the following equations to calculate the genotype frequencies in our population:

p^2 = frequency of red gravel divers,
$2pq$ = frequency of pink gravel divers, and
q^{2}= frequency of white gravel divers.

- ❖ Compare these EXPECTED genotype frequencies to the OBSERVED genotype frequencies from Table 6.3. Are they identical? Provide a biological explanation for these results.

- ❖ After you have finished simulating the effect of predation on your population, get together with the other groups of researchers working on your specific island. Compare your results by considering the following questions:

  a) What changes in genotype frequency did you expect to see in the Northern study site? in the Eastern site? in the Southern site? in the Western site?
  b) Do your observed results agree with your expectations? Be sure that you can explain any deviations between the observed and expected results with a REASONABLE scientific hypothesis.
  c) Assuming that there are no major environmental changes on the island over a long period of time, describe one way in which the predators might adapt to the anticipated distribution of gravel divers on the island. How might these adaptations within the predator population affect the gene pool of the four prey populations on the island?
  d) What would you expect to see if a hurricane hit the island and dumped a large volume of red gravel on the eastern shore of the island?
  e) Can you think of any other "real-life" situations where these types of evolutionary forces have affected the gene pool of a natural population?

Random Genetic Drift

The natives from a distant island have come to your study site. They have decided to take some gravel divers home with them in the hopes of starting a new colony on their home island. As a conscientious ecologist, you spend a great deal of time explaining to them that they should not try to introduce a non-native species such as the gravel diver to their home island. You cite examples of disastrous exotic introductions, such as brown snakes in Hawaii, pigs in the Galapagos, kudzu and zebra mussels in the southeastern U.S., and gypsy moths and Japanese beetles in the continental United States. In spite of all of your best conservation-minded intentions, the natives are "bound and determined" to remove a few gravel divers and bring them home to breed. The night before the natives are scheduled to return home, a small group sneaks down to your study site and removes 6 gravel divers. Unfortunately, they manage to get an equal number of males and females and all of the gravel divers manage to survive and reproduce on the new island. Little do you realize that four years later you will be returning to the new island as you embark on a Ph.D. project to study the impact of the exotic introduction of gravel divers on the native flora and fauna. But for now, let's simulate the effect of this "founding" event. Since the 6 gravel divers that were initially removed were RANDOMLY selected, any variation between the gene pool of the six founding individuals and the original population at your study site is due to *Random Genetic Drift.* In particular, we are simulating a specific example of genetic drift called The Founder Effect. You should be aware that ANY deviation in the gene pool of a population that is due solely to RANDOM events is an example of random genetic drift. In general, this evolutionary agent is only significant in populations with a small number of breeding individuals. The long-term effects of Random Genetic Drift within a single population include increased homozygosity and potential loss of alleles from the population.

❖ *Generating a Hypothesis*: **"**What do you expect to happen at YOUR site?" For each of the following genotype and allele frequencies, clearly indicate whether you expect the frequency to increase (+), decrease (-), or remain the same (0) AFTER genetic drift has occurred at YOUR STUDY SITE:

EXPECTED FREQUENCY CHANGES

	GENOTYPE			*ALLELE*	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (A)	WHITE (a)
Expected Changes (+, -, 0)	________	________	__________	________	_________

❖ Use the sieve to remove a RANDOM sample of gravel divers from your population.

❖ To simulate the night-time conditions, CLOSE YOUR EYES and select SIX of these gravel divers. These six individuals represent the founders that will be brought to the new island and start the new population.

❖ Calculate the allele frequencies for the new founding population as follows:

Frequency of Red ("A") allele:

$$p = \frac{(2)(AA) + Aa}{(2)(N)}$$

Frequency of White ("a") allele:

$$q = 1.0 - p$$

where AA = number of red gravel divers among your 6 founders,
Aa = number of pink gravel divers among your 6 founders,
aa = number of white gravel divers among your 6 founders, and
N = total number of founders (6).

❖ Let us assume that Hardy-Weinberg conditions exist on the new island. Under this assumption, calculate the expected *genotype frequencies* after one generation of random mating. Use the p and q values calculated above:

p^2 = frequency of red gravel divers,
2pq = frequency of pink gravel divers, and
q^2 = frequency of white gravel divers.

❖ Complete Table 6.6 to reflect the effect of random genetic drift. You should note that in this part of the simulation, the random drift occurs prior to mating (when the natives steal a small number of gravel divers). After the small sample of gravel divers has been stolen, they undergo random mating on the new island. So the new population that was founded by the stolen gravel divers is, in fact, in Hardy-Weinberg equilibrium. But any differences that you notice in the gene pool of this new population and the original population from which the founders was stolen is a measure of random genetic drift.

Table 6.6. *Genotype/phenotype and allele frequencies for your gravel diver population before and after gene flow, natural selection, and genetic drift.*

YOUR STUDY SITE:________________________ (Geographical Location)

	GENOTYPE FREQUENCIES			*ALLELE FREQUENCIES*	
	RED (AA)	PINK (Aa)	WHITE (aa)	RED (p) ("A" allele)	WHITE(q) ("A" allele)
Initial Population					
After Gene Flow (*from Table 3*)					
After Natural Selection					
After Genetic Drift (*new island frequencies)*					

❖ *Drawing a Conclusion.* Compare the observed results recorded in the above table with the expected results recorded in the Expected Frequency Changes table. Explain any discrepancies that you saw between the recorded observations and the expected results.

❖ After you have simulated the effect of genetic drift, get together with the groups studying gravel divers living in the other three geographical regions of your island. Compare your results and answer the following questions as a group:

a) When you compare the frequency of the red allele on the new island with the frequency of the red allele at your study site (after predation), does the frequency increase or decrease? Based on the results of the four study groups, would you be able to predict the likelihood of this allele increasing in a second founding event? The answers to these questions should give you some insight as to why this evolutionary agent is called Random genetic drift.
b) Commonly, a founding event is followed by an extensive period of inbreeding. What effect would inbreeding have on the gene pool of the gravel divers on the new island?
c) Suppose that the natives had removed 50 gravel divers from your site (rather than six). Let's assume that these 50 gravel divers survived and reproduced on the new island. What effect would the larger sample size have on your results? Could you predict any shifts in the gene pool due to random genetic drift?

Islands are widely regarded by evolutionary biologists as natural laboratories. Within an island ecosystem, such as the Hawaiian Islands and the Galapagos, it is not at all uncommon to find many endemic species of plants and animals. Now that you have completed the bead simulation part of this lab exercise, discuss the following questions with your fellow island researchers. When everyone has finished this part of the lab, your lab instructor may decide to have all of you get together to discuss the answers to these questions.

a) Reproductive isolation is widely regarded as being essential in the evolution of new species. Discuss the attributes of an island ecosystem that might favor reproductive isolation in general.
b) What types of factors might ensure reproductive isolation among the gravel divers living on the three islands that were studied in this lab? In answering this question, consider both the life history characteristics of the gravel diver and the geographical characteristics of the islands. What types of characteristics would favor speciation? What might be some long-range consequences of this type of reproductive isolation? Would the eventual evolution of multiple species be inevitable? Does the size and location of the island affect the rate and/or occurrence of the speciation process? If so, how?
c) Now consider the four populations of gravel divers living on a single island. Is it at all plausible that these populations might become reproductively isolated? Describe some specific circumstances to support your answer. If reproductive isolation does occur, describe some long-range consequences that you might expect to observe in the gravel diver populations. If speciation

were to occur on a single island, what types of adaptations might you expect to evolve in the GDP predator population?

Computer Simulation

Name of Software: Evolving: Examples of the Effects of Natural Selection (Version 1.0)
Source of Software: BioSim Productions
8501 Manastash Road
Ellensburg, WA 98926
http://www.ellensburg.com/~biosim
e-mail: biosim@ellensburg.com
phone: (509) 925-4715

The peppered moth (*Biston betularia*) was studied in England during the late 1800's by Kettlewell and represents a classical study of natural selection in action. Among this particular species of moth, there are two different phenotypes: the peppered (black and white) moth and the all black moth. In these moths, the black allele is completely dominant over the peppered allele (Black moths =BB or Bb, Peppered moths = bb). These moths commonly spend much of their adult life resting on tree trunks trying to avoid being eaten by birds. Prior to the late 1800's, the peppered phenotype of this moth was most commonly found living on tree trunks in England.

The climate in England is cool and moist and favors the growth of a symbiotic organism called a lichen on tree trunks. In a symbiotic relationship, two organisms (in this case an algae and a fungus) live together in a mutually beneficial relationship. The lichen growing on most English tree trunks has a greenish-white appearance. Tree trunks that support lichens have a mottled black/greenish-white appearance. So you can imagine that the peppered moth would be afforded a great deal of camouflage from the predatory birds if it lived among the lichens on such a tree trunk. Lichens are severely affected by pollution and commonly die off as soon as the environmental quality declines—kind of like the canary in a mine!

Along comes the Industrial Revolution. In the absence of any EPA regulations, the air in English cities, such as Birmingham, becomes very polluted. The lichens die leaving very dark, black tree trunks. The peppered form of the moth is no longer camouflaged and the predatory birds fill themselves on moths. Suddenly, it becomes most advantageous to be a black form of the moth. The environmental changes associated with the Industrial Revolution have changed the direction of natural selection as it operates on this particular population.

In the second part of this lab, you will simulate these evolutionary changes in the peppered moth population using the Evolving computer software program. Your lab instructor has already booted up the Evolving software program at the computers in the lab. To perform this computer simulation, you should follow these instructions:

- Using the Evolving software, choose the peppered moth simulation.
- Choose the following options within this simulation program:
    - a) not polluted
    - b) phenotypes equally frequent

- Quickly eat about 20 moths in each of three generations
  a) What happens to the moth population over the three year period? (Select the Option key on the main menu and look at your data both in a tabular and a graphical form). You should write down the allele frequency for one of the alleles in each generation.
  b) Did you eliminate one of the alleles? If not, continue eating 20 moths/generation until you do.
  c) How many generations did it take to eliminate one of the alleles?
- Repeat the above simulation in a highly polluted environment. Continue until you have again eliminated one of the alleles.
  a) How many generations did it take to eliminate an allele this time?
  b) Considering the outcome of the two studies, what generalization can you make about which allele is most beneficial (and therefore provides greater fitness)?
  c) Why was it more difficult to eliminate an allele in one of the two simulations? (Hint: think about the genetic basis for this trait.)

Appendix

Order information for the plastic beads:

We purchased the beads through the local Wal-Mart store. They are distributed by the following company:

The Beadery Craft Products
P.O. Box 178
Hope Valley, RI 02832

The specific order information is as follows:
Item #697A: 4 mm Faceted Beads, 200 count (cost: $0.58/bag of 200 beads)
Item #825A: 11 mm Tri Beads, 145 count (cost: $0.58/bag of 145 beads)

Color Codes for both sets of beads:

006: Crystal (White or Clear)
015: Pink
013: Ruby (Red)
007: Emerald (Green)

Beads may also be ordered from:

Bolek's Craft Supplies
330 N. Tuscarawas Ave
Dover, OH 44622
e-mail: www.boleckscrafts.com

Construction specifications for sieve:

The size difference between the faceted and the tri beads is critical since it allows for easy separation of the beads using the mesh sieve. Specifically, the sieve size is small enough to allow the faceted beads to pass through the sieve while capturing the tri beads. To make a sieve, cut out the bottom of plastic food containers (ex. large butter containers, ricotta cheese containers, etc.) and insert a piece of 1/4 inch mesh hardware cloth. The hardware cloth is both glued and sewed in place using nylon fishing line. It is important to ensure that beads will not be trapped between the edge of the hardware cloth and the plastic container. The primary purpose of the sieve is to allow for easy separation of the two types of beads at the end of the lab. At the end of the lab exercise, we have the students sieve through the faceted beads and remove all of the tri beads. In this way, there should be no additional set-up for the next lab section.