LABORATORY 8. POPULATION GENETICS AND EVOLUTION

Objectives

In this activity, you will

- learn about the Hardy-Weinberg law of genetic equilibrium
- study the relationship between evolution and changes in allele frequency by using your class as a sample population

Required Knowledge

Before beginning this laboratory, you should understand

- the process of meiosis and its relationship to the segregation of alleles
- the basics of Mendelian Genetics
- the Hardy-Weinberg equation and its use in determining the frequency of alleles in a population
- that natural selection can alter allelic frequencies in a population
- the effects of allelic frequencies of selection against the homozygous recessive or other genotypes

Expectations

At the completion of this laboratory, you should be able to

- calculate the frequencies of alleles and genotypes in the gene pool of a population using the Hardy-Weinberg formula
- discuss natural selection and other causes of microevolution as deviations from the conditions required to maintain Hardy-Weinberg equilibrium

Background

The **Hardy-Weinberg Theorem** states that the frequencies of alleles in a sexually reproducing population remain constant (in equilibrium) from generation to generation unless acted upon by outside factors. That is, if we consider two alleles, *A* and *a*, in a population, the reshuffling of alleles that occurs due to meiosis and recombination does not change the numbers of these alleles in the population. Hardy and Weinberg argued that a population's allele and genotype frequencies would remain statistically constant as long as five conditions were met:

1. **The breeding population is very large.** In a small population, chance events can greatly alter allele frequency. For example, if only five individuals of a small population of deer carry allele *a* and none of the five reproduce, allele *a* is eliminated from the population.
2. **Mating is random.** Individuals show no preference for a particular phenotype.
3. **There is no mutation of the alleles.**
4. **No differential migration occurs.** No immigration or emigration.
5. **There is no selection.** All genotypes have an equal chance of surviving and reproducing.

The Hardy-Weinberg Theorem provides a yardstick by which we can measure changes in allele frequency, and therefore, in evolution. If we can determine the frequency of a pair of alleles in a population, we can sample that population over several generations and answer the question, "Is the population evolving with respect to these particular alleles?" The **Hardy-Weinberg equations** can be applied to estimate the frequencies of specified alleles within a population at any given time.

Consider the following data on Rh blood type from a hypothetical human population:

Table 8.1 Results of Testing for Human Rh Blood Type

Blood Type	Number of People
Rh+	4,680
Rh–	1,320
Total	6,000

Assume that Rh blood type is inherited through two alleles: *D*, a dominant allele for Rh+ blood type, and a recessive allele *d* for Rh– blood type.

Using the Hardy-Weinberg equations,

let p = frequency of the dominant allele (*D*, in this case)

and q = frequency of the recessive allele (*d*)

Each person is either Rh+ or Rh–

therefore,

$p + q = 1$ or 100% of the population

Since humans are diploid, individuals may be homozygous dominant (*D/D*), heterozygous (*D/d*), or homozygous recessive (*d/d*). Thus, the basic equation must be expanded to represent all the genotype frequencies.

Thus, $(p + q)(p + q) = 1$

or $p^2 + 2pq + q^2 = 1$

where p^2 = frequency of the homozygous dominant (*D/D*),

$2pq$ = frequency of the heterozygous condition (*D/d*),

and q^2 = frequency of the homozygous recessive (*d/d*).

Notice that $p^2 + 2pq$ = frequency of the Rh+ phenotype in the population.

From the data in Table 8.1, we can now calculate q^2, the frequency of the homozygous recessive:

$q^2 = 1320/6000 = 0.22$

then $q = \sqrt{0.22} = 0.47$

$p + q = 1$

$p = 1 - q$

$p = 1 - 0.47 = 0.53$

This tells us that 53% of the population tested has the allele *D* and 47% has the allele *d*.

If we repeated our sample over several generations, we could tell if this population is evolving with respect to the Rh factor alleles.

Notice that we had to calculate a value for q before we could we could determine p. Why is this true?

Exercise 8A: Estimating Allele Frequencies for a Specific Trait Within a Sample Population

Introduction

Using the class as a sample population, you will estimate the allele frequency of a gene controlling the ability to taste the chemical PTC (phenylthiocarbamide). A bitter-taste reaction to PTC is evidence of the presence of a *dominant allele* in either the homozygous condition (*A/A*) or the heterozygous condition (*A/a*). The inability to taste the chemical at all depends on the presence of homozygous recessive alleles (*a/a*).

Procedure

1. Press a strip of control taste paper to your tongue tip. Wait until your saliva has completely saturated the paper, then note the taste of the paper.
2. Now repeat Step 1 using the PTC taste paper. PTC tasters will sense a bitter taste. For the purposes of this exercise, these individuals are considered to be tasters. If you sense little more than the taste of the paper itself, you are a nontaster. Record the class data in Table 8.2.

Questions

1. Use your class data and the Hardy-Weinberg equations to complete Table 8.2.

Table 8.2 Phenotypic Proportions of Tasters and Nontasters and Frequencies of the Determining Alleles

	Phenotypes					
	Tasters ($p^2 + 2pq$)		Nontasters (q^2)		Frequency of the Alleles (%)	
	Count	% of Total	Count	% of Total	p	q
Class Data						
North American Population		70		30		

2. What percentage of your class are heterozygous tasters? _____

3. What are some reasons that the values for p and q for your class might differ from the same frequencies reported for the North American population?

4. Would you expect the frequencies of the alleles for PTC tasting and nontasting to remain constant for North America over the next 200 years? State your answer in terms of the five conditions that Hardy and Weinberg stated must be fulfilled for allele frequencies to remain constant.

Exercise 8B: A Test of Hardy-Weinberg Equilibrium

Introduction

In this activity, your entire class will simulate a population of randomly mating individuals. Choose another student at random (for this simulation, assume that gender and genotype are irrelevant to mate selection.) The population begins with a frequency of 0.5 (50%) for the dominant allele *A* and also 0.5 (50%) for the recessive allele *a*. Your initial genotype is *A/a*. Record this on the Data Sheet. You have four cards: each represents a chromosome. Two cards (chromosomes) will have allele *A* and two cards will have allele *a*. The four cards represent the products of meiosis. Each "parent" will contribute a haploid set of chromosomes to the next generation.

Procedure

1. Turn the four cards over so the letters are not showing, shuffle them, and take the card on top to contribute to the production of the first offspring. Your partner should do the same. Put the two cards together. The two cards represent the alleles of the first offspring. One of you should record the genotype of this offspring as the Generation 1 Genotype on his or her Data Sheet.

2. Retrieve your cards and reshuffle them. Repeat Step 1 to produce a second offspring. The second partner records the genotype of this offspring on his or her Data Sheet. The very short reproductive career of this generation is over.

3. You and your partner now become the next generation by assuming the genotypes of the two offspring you have produced. That is, Student 1 assumes the genotype of the first offspring and Student 2 assumes the genotype of the second offspring as you have recorded them on your Data Sheets. Obtain additional cards if necessary. For example, if you now have the genotype *a/a*, you will need four cards, all marked *a*. If you have the genotype *A/A*, you will need four cards all marked *A*. If you have the genotype *A/a*, keep the original four cards.

4. Now, **randomly** seek out another person with whom to mate in order to produce the offspring of the next generation. The sex of your mate does not matter, nor does the genotype. Repeat Steps 1–3, being sure to record your new genotype, after each generation, on your Data Sheet. Repeat this exercise to produce five generations.

5. Your teacher will collect class data for Generation 5 by asking you to raise your hand to report your genotype. Record the class totals in Table 8.3

Table 8.3 Class Totals for Exercise 8B

	Genotype Totals		
Generation	***A/A***	***A/a***	***a/a***
1			
2			
3			
4			
5			

Data Analysis

Compare your data for Generation 5 in Table 8.3 with the class data for PTC tasting in Table 8.2. What information do you have for Generation 5 that you do not have in Table 8.2?

From Table 8.3, what is the population size? _____

The Hardy-Weinberg equations are used to give estimates of allele frequencies for large populations. Here, we have a small population and there is no need to estimate allele frequencies, because we have actual counts. Calculate p and q as follows:

Number of *A* alleles present at the fifth generation

Number of offspring with genotype *A/A* _____ × 2 = _____ *A* alleles

Number of offspring with genotype *A/a* _____ × 1 = _____ *A* alleles

Total = _____ *A* alleles

$$p = \frac{\text{Total number of } A \text{ alleles}}{\text{Total number of alleles in the population}} = \_\_\_\_\_$$

In this case, the total number of alleles in the population is equal to the number of students in the class × 2.

Number of *a* alleles present at the fifth generation

Number of offspring with genotype *a/a* _____ × 2 = _____ *a* alleles

Number of offspring with genotype *A/a* _____ × 1 = _____ *a* alleles

Total = _____ *a* alleles

$$q = \frac{\text{Total number of } a \text{ alleles}}{\text{Total number of alleles in the population}} = \_\_\_\_\_$$

Questions

1. What are the frequencies of the alleles in Generation 5?
   a. the frequency (q) of allele *a* in Generation 5? _____
   b. the frequency (p) of allele *A* in Generation 5? _____
2. Are the values for p and q in Generation 5 different from the beginning values?
   _____
3. Is your answer to Question 1, above, consistent with the alleles being at equilibrium?
   _____
   If not, why not?
   _____
   _____
   _____
   _____
   _____
   _____

4. Look back at the five conditions that must be met for allele frequencies to remain constant. Which, if any, of these conditions might not have been met in this simulation?

Exercise 8C: Selection

Introduction

In nature, not all genotypes have the same rate of survival; that is, the environment might favor some genotypes while selecting against others. An example is human sickle-cell anemia, a disease caused by a single gene mutation. Individuals who are homozygous recessive (*a/a*) often do not survive to reach reproductive maturity. In this simulation, you will assume that the homozygous recessive individuals never survive (100% selection against), and that heterozygous and homozygous dominant individuals survive 100% of the time.

Procedure

Everyone begins with the heterozygous genotype; thus, the initial frequency of each of the alleles is again 0.5 (50%). Follow the procedure in Exercise 8B, with the following modification: every time your "offspring" is *a/a*, assume that it does not survive to reproduce. Because you want to maintain a constant population size, the same two parents must try again until they produce two surviving offspring.

Proceed through five generations, selecting against the homozygous recessive offspring 100% of the time. Then, calculate the new p and q frequencies using the method from 8B.

Table 8.4 Class Totals for Exercise 8C

	Genotype Totals		
Generation	***A/A***	***A/a***	***a/a***
1			
2			
3			
4			
5			

Questions

1. What are the frequencies of the alleles in Generation 5?

   a. the frequency (q) of allele *a* in Generation 5? _____

   b. the frequency (p) of allele *A* in Generation 5? _____

2. Are the values for p and q in Generation 5 different from the beginning values? Explain your answer.

3. Compare the values for p and q calculated for Generation 5, Exercise 8B with the values you just calculated for Exercise 8C. How do the new frequencies of p and q compare to the initial frequencies in Exercise 8B?

4. Predict what would happen to the frequencies of p and q if you simulated another five generations.

5. In a large population, would it be possible to completely eliminate a deleterious recessive allele? Explain your answer.

Exercise 8D: Heterozygote Advantage

Introduction

From Exercise 8C, it is easy to see that the lethal recessive allele rapidly decreases in the population. However, studies show an unexpectedly high frequency of the sickle-cell allele in some human populations. These populations exist in areas where malaria is (or until recently was) killing many people. It seems that individuals who are heterozygous for sickle-cell anemia are slightly more resistant to a deadly form of malaria than are homozygous dominant individuals. In malaria-ridden areas, there is a slight selection against homozygous dominant individuals as compared to heterozygotes. This fact is easily incorporated into our simulations.

Procedure

Begin again with the genotype *A/a*. Follow the procedure in Exercise 8B, with the following modifications: if your offspring is *A/A*, flip a coin. If heads, the offspring does not survive. If tails, the offspring does survive. The genotype a/a never survives. Parents must produce two surviving offspring each generation. This time, simulate ten generations. Total the class genotypes and then calculate the *p* and *q* frequencies for Generation 5 and for Generation 10. If time permits, the results from another five generations would be extremely informative.

Table 8.5 Class Totals for Exercise 8D

	Genotype Totals		
Generation	***A/A***	***A/a***	***a/a***
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			

Questions

1. What are the frequencies of the alleles in Generation 10?
   a. the frequency (*q*) of allele *a* in Generation 10? _____
   b. the frequency (*p*) of allele *A* in Generation 10? _____
2. Account for the differences in *p* and *q* frequencies from Exercise 8C to Exercise 8D.

   _____

   _____

   _____

   _____

   _____

   _____
3. Do you think the recessive allele will be completely eliminated in either Exercise 8C or Exercise 8D? Explain your answer.

   _____

   _____

   _____

   _____

   _____

   _____

4. What is the importance of heterozygotes (the heterozygote advantage) in maintaining genetic variation in populations?

5. Suppose you repeated this activity, but you did the coin toss to determine if the *A/a* individuals reproduce and all of the *A/A* individuals reproduced. How would you expect this to change the allele frequencies for Generation 10?

Exercise 8E: Genetic Drift

Introduction

It is possible to use our simulation to look at the phenomenon of genetic drift in detail.

Procedure

Divide the lab into several smaller, isolated populations. For example, a class of 30 could be divided into 3 separate populations of 10 individuals each. Individuals from one population do not interact with individuals from other populations. Follow the procedure in Exercise 8B through five generations. Record the new genotypic frequencies and then calculate the new frequencies of p and q for each population.

Questions

1. Explain how the initial genotypic frequencies of the populations compare.

2. What do your results indicate about the importance of population size as an evolutionary force?

Hardy-Weinberg Problems

1. In *Drosophila*, the allele for normal length wings is dominant over the allele for vestigial wings (vestigial wings are stubby, little curls that cannot be used for flight). In a population of 1,000 individuals, 360 show the recessive phenotype. Use the Hardy-Weinberg equations to estimate the number of homozygous dominant and heterozygous individuals.

2. The allele for the ability to roll the tongue into a tube is dominant over the allele for the lack of this ability. In a population of 500 individuals, 25% show the recessive phenotype. Use the Hardy-Weinberg equations to estimate the number of homozygous dominant and heterozygous individuals.

3. The allele for the hair pattern called "widow's peak" is dominant over the allele for no widow's peak. In a population of 1,000 individuals, 510 show the dominant phenotype. About how many individuals would have each one of the three possible genotypes?

4. In the United States, about 16% of the population is Rh–. The allele for Rh– is recessive to the allele for Rh+. If a high school has a population of 2,000 students, about how many students would have each one of the three possible genotypes?

5. In certain African countries, 4% of the newborn babies have sickle-cell anemia, which is a recessive trait. Out of a random population of 1,000 newborn babies, about how many babies would have each one of the three possible genotypes?

6. In a certain population, the dominant phenotype of a certain trait occurs 91% of the time. What is the frequency of the dominant allele?

Name _____

LABORATORY 8 **DATA SHEET** **Date** _____

Exercise 8B
Hardy-Weinberg Equilibrium
Initial Class Frequencies
***p* = 0.5** ***q* = 0.5**

My Initial Genotype	*A/a*
Generation 1 Genotype	
Generation 2 Genotype	
Generation 3 Genotype	
Generation 4 Genotype	
Generation 5 Genotype	
Final Class Frequencies	
p =	*q* =

Exercise 8C
Selection
Initial Class Frequencies
***p* = 0.5** ***q* = 0.5**

My Initial Genotype	*A/a*
Generation 1 Genotype	
Generation 2 Genotype	
Generation 3 Genotype	
Generation 4 Genotype	
Generation 5 Genotype	
Final Class Frequencies	
p =	*q* =

Exercise 8D
Heterozygote Advantage
Initial Class Frequencies
***p* = 0.5** ***q* = 0.5**

My Initial Genotype	*A/a*
Generation 1 Genotype	
Generation 2 Genotype	
Generation 3 Genotype	
Generation 4 Genotype	
Generation 5 Genotype	
Final Class Frequencies, Generation 5	
p =	*q* =

Generation 6 Genotype	
Generation 7 Genotype	
Generation 8 Genotype	
Generation 9 Genotype	
Generation 10 Genotype	
Final Class Frequencies, Generation 6	
p =	*q* =

Exercise 8E
Genetic Drift
Initial Class Frequencies
***p* = 0.5** ***q* = 0.5**

My Initial Genotype	*A/a*
Generation 1 Genotype	
Generation 2 Genotype	
Generation 3 Genotype	
Generation 4 Genotype	
Generation 5 Genotype	
Final Class Frequencies	
p =	*q* =