Battle of the Bugs: Pesticide Resistance

KEY CONCEPT:

Pesticide resistance evolves through the process of natural selection. More specifically, it is a form of artificial selection driven by the actions of humans. It is important for humans to find new and effective ways to manage pests because of this rapid evolution of resistance to pesticides.

OBJECTIVES:

Students will be able to:

- Understand and explain how pesticide resistance evolves through the process of natural selection.
- Understand how pesticide resistance is affecting pest management in bed bugs.

BACKGROUND BASICS:

We often try to control our environment, especially when it comes to things we consider to be pests. We consider things to be pests because they eat our plants, transmit diseases to humans or other animals, invade our homes, or are just annoying to people, their pets, or to livestock. One way that humans control pests is through the use of **pesticides**, or chemicals that are formulated to kill pests. You may think this is a modern practice, but there are ancient texts from 2500 years ago in Samaria that refer to using sulfur to kill insects. By the tenth century, scholars had even created a collection of agricultural practices, called Geoponika, that included pest control methods.

If you read information from the World Health Organization (WHO) and the U.S. Centers for Disease Control and Prevention (CDC) websites, you will find that pesticides have benefited humans in many ways, especially in mosquito control. We often hear of the harmful impacts of pesticides, and forget of their benefits. Pesticides can reduce the number of people who get sick or die from diseases that are transmitted by pests, improve the quality of our food, increase the productivity of our crops, and reduce damage to human homes and belongings. However, pesticides are not without their own problems. Pesticides, even natural pesticides, are designed to kill pests. When not used properly or judiciously, they may have side-effects. Some of the harmful effects of pesticides include the potential to kill beneficial organisms that help control pest populations, affecting non-target organisms such as birds, fish, and other wildlife, having adverse environmental effects, or causing dangers to human health. One other concern is the potential for pesticide resistance to evolve within pest populations. Over the past 150 years, humans have become more and more dependent on pesticides to manage pests. In 1914, pesticide resistance began to be documented. That means that when a certain amount of pesticides were applied, they were no longer killing the pests they once killed.

LEARNING CONCEPT:

- Environmental understanding
- Critical thinking

GRADE LEVEL: 7

SUBJECT: Science

SETTING: Indoors; classroom with desks or tables

TIME NEEDED: 1-2 class periods

ENTOMOLOGY SKILL: Understanding how natural selection can lead to pesticide resistance in insects.

LIFE SKILL: Communication, critical thinking

SUNSHINE STATE STANDARDS: SC.7.N.1.1, SC.7.L.15.2, SC.7.L.16.1, SC.7.L.16.2

MATERIALS NEEDED:

Copies of article "The History of Bed Bug Management—with lessons from the past" by Michael F. Potter (see "Activity Introduction" for more information)

Genotype cards for simulation activity

Copies of student worksheets included in lesson

Pens/pencils

Farmers would need to use more or different pesticides to kill the same population of pests. This led to something called the “pesticide treadmill.” To reduce cost of pest management and reduce environmental impacts, farmers began to go back to the basics and use an integrated approach to pest management, integrated pest management (IPM). In IPM, they rotated different classes of pesticides, used better farming practices, such as plowing under stalks so pests could not survive the winter, and monitored for pest activity so pesticides could be used more effectively.

What is pesticide resistance?
Pesticide resistance is the genetically inherited ability of an organism to survive a pesticide application. This occurs when a pesticide is applied to a given pest population where within that population there are some pests with the genetic ability to survive the pesticide application. Those pests with this genetic ability to survive the pesticide may then reproduce, producing more offspring that inherit resistance to the pesticide, and so on until the population is almost entirely resistant to the pesticide. Since insects have high reproductive rates, resistance in a pest population can happen in a short period of time.

How does pesticide resistance occur?
Pesticide resistance occurs through the process of evolution by natural selection. In this case, humans artificially drive the selection by applying pesticides. An understanding of natural selection is key to understanding pesticide resistance within a given population.

Biological evolution is simply descent with modification, which includes **microevolution** (changes in gene frequencies within a population over time) and **macroevolution** (descent of different species from a common ancestor over a long period of time) (“Understanding Evolution: An Introduction to Evolution” 2012). These changes are responsible for the amazing diversity of life we see on our planet today. The mechanisms by which evolution can occur include **mutation** (changes in DNA that occur during DNA replication or due to external sources, such as radiation), **gene flow/migration** (change to gene frequencies that is due to the movement of individuals in or out of a population), **genetic drift** (random changes to a population over time), **sexual selection** (selection that occurs through competition for mates or from mate choice), and **natural selection** (Futuyma 2009; “Understanding Evolution: Mechanisms: the Process of Evolution” 2012).

Charles Darwin defined natural selection as the “differential survival and reproduction of organisms with genetic characteristics, enabling them to better use environmental resources.” When you think about that, you see that characteristics must be heritable and there must be variations in the population that give some individuals a better chance of survival and reproduction than others. Variation in a gene pool can occur through mutations, gene flow, or sexual reproduction. Those variations must be able to benefit the individual to make them better able to survive and reproduce. Not only that, but the variation must be heritable, or made of genetic information that can be passed from one generation to the next. So in review, there must be **variability**, **heritability,** and **differential reproductive success** for natural selection to take place in a population. Over-time, characteristics (frequencies of alleles) will change as those with certain traits that make them better-suited to the environment leave more offspring and those not so well suited leave fewer offspring.

Now, back to pesticide resistance. When a pesticide is applied to a given pest population, there may be **genetic variation** in the population where some individuals may confer the ability to survive the pesticide, while other individuals are killed by the pesticide, based upon their underlying genotypes. There is **differential reproductive success**, because those with resistance to the pesticide survive and reproduce in greater numbers than those not resistant to the pesticide.

This trait is **heritable** because it is due to the genotype and therefore, a number of the offspring of the survivors inherit resistance to the pesticide. These resistant pests survive the next pesticide application, and produce more offspring that inherit resistance to the pesticide. If a similar pesticide is applied frequently, the resistant pests will soon make up most of the pest population and the pesticide will no longer work as a method of control.

What can be done to slow pesticide resistance?
There are several ways we can slow resistance to pesticides. One of the best methods would be to use Integrated Pest Management (see IPM lesson for more information about this topic). IPM uses a variety of management techniques including, education about the pest, identification, monitoring, sanitation, cultural practices, biological controls, and target pesticide applications. However, when pesticides are necessary, the classes of pesticides are rotated and they are only used when pests are present and in a targeted fashion so the environmental impact is limited. Pesticides are classified according to how they work against the insect. Some work against various parts of the insect's nervous system while others work against the insect's digestive system, respiratory system, or endocrine system. Rotating between the different classes of pesticides reduces the chances that the insects will become resistant to the pesticides.

Teacher's Note: *Gregor Mendel, Pea Plants, and Genetics*

The work of Gregor Mendel was key to understanding heredity in organisms. Prior to Mendel's work, it was thought that offspring inherited blended characteristics from their parents. Through his cross-breeding experiments with pea plants, Gregor Mendel determined that this earlier hypothesis was incorrect by showing that seven different traits in pea plants never displayed intermediate forms.
Mendel's conclusion from his experiments are summarized in two basic principles of heredity:

1. **Principle of segregation**—two alleles for a particular character separate during gamete formation and end up in different gametes.
2. **Principle of independent assortment**—each pair of alleles separates independently of each other pair of alleles during gamete formation.

While today we know that there are exceptions to these rules (e.g., gene linkage), Mendel's work was still critically important to the beginnings of modern genetics. And although Mendel worked with pea plants, the basic underlying principles of heredity are applicable to other organisms, including insects and other invertebrates, birds, reptiles, amphibians, and mammals. (Campbell *et al*. 2008; O'Neil 2013)

ACTIVITY INTRODUCTION:

Pesticide Resistance and Bed Bugs
Bed bug infestations were very common in the United States prior to World War II. But, bed bugs were nearly eradicated during the 1940's and 1950's, largely because of the introduction and widespread availability of DDT to consumers. After several decades of relief from bed bugs, they have recently made a comeback and are being found in large numbers, especially in urban areas, such as homes, schools, hotels, public transportation, offices, movie theaters, and other areas. There are many potential reasons for this resurgence, including increased international travel, decreased vigilance, and increasing pesticide resistance in bed bug populations.

Pesticide resistance seems to be a large culprit; it has been found that bed bugs are now mostly resistant to the pesticides that were most commonly used and were previously most effective to control them. For a brief history of bed bug control, watch Part 1: History of Bed Bug Control in the "History of Bed Bug Management" video located at http://njaes.rutgers.edu/bedbug/?videos#professionals.

Let's Do:

1. Ask students to read the article "The History of Bed Bug Management—with Lessons from the Past" by Michael F. Potter. This article is available at http://entsoc.org/PDF/2011/AE-Potter-spring2011.pdf. Discuss the article with the students and ask them the following questions:

When were bed bugs first treated with modern insecticides?

Bed bugs were first treated with modern insecticides in the 1800's.

When did resistance to insecticides first appear?

Reports of resistance to DDT were first noted in 1947, only a few years after DDT first began to be used. By the mid 1950s, bed bugs were widely resistant to DDT.

Are bed bugs still resistant to insecticides?

Yes. It has been determined that bed bugs have evolved resistance to many pesticide ` *classes. Studies have suggested that some chemicals are still effective in controlling bed bugs, however, "history has shown that bed bugs can develop resistance to insecticides at a rapid rate."* (Potter 2011)

2. To demonstrate how resistance occurs and increases within a population, complete the Bed Bug Resistance Simulation activity with the students as a class as according to the instructions that follow. Ask students to complete the Punnett Square worksheet to determine the resulting genotypes and phenotypes from each set of mating pairs before starting the simulation.
3. Have students complete the follow-up questions on the Bed Bug Resistance Simulation Activity worksheet.
4. Discuss students' answers to the worksheet as well as the questions found in the "Let's Reflect" and "Let's Apply" sections.

Let's Reflect:

What are some of the benefits to using pesticides? What are some of the problems?

Some of the benefits to using pesticides include reducing the number of people who get sick or die from diseases that are transmitted by pests, improving the quality of our food and increasing productivity of our crops, and reducing damage to human homes and belongings. Some of the problems with using pesticides are that pesticides have the potential to kill beneficial organisms that help control pest populations, they may affect non-target organisms such as birds, fish, and other wildlife, they can have adverse environmental effects, they may cause dangers to human health, and pesticide resistance may evolve within pest populations.

How does pesticide resistance occur? Listen for key words like variation, inheritance/heritability, and differential reproductive success.

When a pesticide is applied to a given pest population, there may be genetic variation in the population where some individuals may confer the ability to survive the pesticide, while other individuals are killed by the pesticide, based upon their underlying genotypes.

There is differential reproductive success, because those with resistance to the pesticide survive and reproduce in greater numbers than those not resistant to the pesticide. This trait is heritable (genotype) and therefore, a number of the offspring of the survivors inherit resistance to the pesticide. These resistant pests survive the next pesticide application, and produce more offspring that inherit resistance to the pesticide and so on until the population is almost entirely resistant to the pesticide. The resistant pests will soon make up most of the pest population and the pesticide will no longer work as a method of control.

What happened in your simulation? Was the last generation resistant, non-resistant, or a mix of both? What do you think would happen if you continued the simulation for several more generations? This simulation could potentially have different results every time it is done. Why do you think this is? What factors do you think affect how a population might evolve pesticide resistance in "real life?" Is the simulation realistic? Why or why not?

The factors that could affect the results of the simulation include the genotypes of the individuals of the starting population and the random selection of mates within each generation (and therefore, the genotypes of the resulting offspring). In real life, the factors could include the genotypes and size of the starting population, random mating and resulting genotypes of the offspring, the number of offspring produced (reproductive success), selection pressures (e.g., number of times and the types of pesticides sprayed within a population), mutations, and so on. Also, the simulation is not necessarily true to what would happen in real life, because in real life the starting population would likely be bigger than two individuals and therefore might have more genetic variation, the number of offspring would vary, the parents would not necessarily die after mating and would likely mate multiple times with different partners, mate selection may not be random, mutations could occur, pesticide resistance might be controlled through the dominant allele (instead of recessive as in the simulation), and pesticide resistance might be under the control of multiple genes (instead of just one as indicated in the simulation activity).

Let's Apply:

How do you think pesticide resistance occurred in bed bugs? Should we keep looking for new pesticides that will work instead of the ones we previously used? Why or why not?

When a particular pesticide (or class of pesticides) was applied to a population of bed bugs, there was genetic variation in the population where some of the bed bugs had the ability to survive the pesticide, while other individuals were killed by the pesticide, based upon their underlying genotypes. Those bed bugs with resistance to the pesticide survived and reproduced in greater numbers than those bed bugs not resistant to the pesticide. The offspring of the survivors could inherit resistance to the pesticide. These resistant bed bugs survive the next pesticide application, and produce more bed bugs that inherit resistance to the pesticide, and so on until the population is mostly (or completely) comprised of bed bugs resistant to the pesticide.

While looking for new pesticides might appear advantageous, it might be better to look for other methods to control bed bugs that would be better for the environment, pose less risk to human health and the environment, and have reduced likelihood for resistance to evolve. Also, it might be useful to find combinations of control methods (this is called Integrated Pest Management—see the "Bed Bug IPM" lesson for more information) that work well for controlling bed bugs.

What other methods could you use (instead of pesticides) to control for bed bugs?

Bed bugs are susceptible to heat. Bed bugs will die at 120℉. Since clothes dryers reach this temperature, linens and clothing can be dried on high heat for at least 1 hour (Note: Dryers have a heat -up and cool-down cycle, so 1 hour is recommended. It only takes a minute or so at a high temperature to kill a bed bug.) Since bed bugs are attracted to humans to feed, climb up traps can be used to prevent bed bugs from climbing up on furniture. Please visit http://news.ufl.edu/2014/05/20/homemade-bedbug-trap/ on tips on how to make a bed bug trap out of recyclable materials. Holes or cracks in building structures and behind baseboards should be fixed or sealed to reduce bed bug harborages, and bed bugs can be vacuumed up (Note that vacuuming may leave behind most eggs). Since heat will kill bed bugs, trained pest management specialists can heat and/or steam-treat areas to kill bed bugs. Heat and low pressure steam are a good way to treat bed frames , mattresses,, and box springs. After a treatment, a mattress encasement may be installed to prevent bed bugs from harboring in the seams of the mattress.

EXTENSION:

Have students use the internet to find and read a news article about the evolution of pesticide resistance in bed bugs and/or about how bed bug infestations are becoming more common in the United States. Ask your students to write their reflections about the news article. Also, have them look for the vocabulary words discussed in this section (evolution, natural selection, variation, heritability/inheritance, reproductive success, DNA, gene, allele, mutation, genotype, phenotype, homozygous, heterozygous, dominant allele, recessive allele, etc.) and discuss how it relates to pesticide resistance in bed bugs in correspondence with the particular article they read. One interesting article, "Bed Bugs Bite Back Thanks to Evolution," can be found on the Understanding Evolution website at http://evolution.berkeley.edu/evolibrary/search/lessonsummary.php?&thisaudience=9-12&resource_id=418. This resource also includes a video podcast, additional resources and articles, and possible discussion questions.

Are there natural remedies to bed bugs? Have students read the article "How a Leafy Folk Remedy Stopped Bed Bugs in their Tracks," which can be found at http://www.nytimes.com/2013/04/10/science/earth/how-a-leafy-folk-remedy-stopped-bedbugs-in-their-tracks.html?_r=1&. Summarize the findings discussed in this article to students and show the students the photographs and video clip included with the article. Ask students to write a response (either individually or in small groups) to the following question:

Could bed bugs develop resistance to the leaves? Why or why not?

Yes, they could develop resistance to the leaves. If there was genetic variation in the population where some of the bed bugs had the ability to survive the hooks on the leaves while other individuals were killed by the hooks. Those bed bugs with resistance to the hooks would need to survive and reproduce in greater numbers than those bed bugs not resistant to the hooks, then eventually resistance to the hooks on the leaves could evolve. For example, if some individuals had a thicker exoskeleton where their legs bend, they would not be pierced by the hooks. Those thick-skinned individuals could pass this trait to their offspring. If the population was continually exposed to the leaves, eventually all the population would have thick skin and would be able to survive.

VOCABULARY

Pesticide: The Food and Agriculture Organization (FAO) has defined a pesticide as "any substance or mixture of substances intended for preventing, destroying, or controlling any pest, including vectors of human or animal disease, unwanted species of plants or animals, causing harm during or otherwise interfering with the production, processing, storage, transport, or marketing of food, agricultural commodities, wood and wood products or animal feedstuffs, or substances that may be administered to animals for the control of insects, arachnids, or other pests in or on their bodies. The term includes substances intended for use as a plant growth regulator, defoliant, desiccant, or agent for thinning fruit or preventing the premature fall of fruit. Also used as substances applied to crops either before or after harvest to protect the commodity from deterioration during storage and transport." http://www.fao.org/docrep/005/y2200e/y2200e07.htm

Pesticide class: Pesticides can be grouped by target organism or by chemical family. When a pesticide is grouped by its chemistry, scientists use the mode of action, or MOA, for grouping. The mode of action is how the pesticide affects the target organism at the tissue or cellular level. For example, a pyrethroid is a chemical class of synthetic insecticide that is based on the natural permethrin that is found in the chrysanthemum flower. It works on the insect nervous system, specifically on the sodium channel, and is found in common products like Raid. When people want to slow resistance in an insect population, they rotate the pesticides at the class level. For more information about pesticide classes, please visit: http://www.irac-online.org/documents/moa-classification/?ext=pdf.

Species: a group of individuals that can (actually or potentially) interbreed. (Note: the definition of a species has long been under debate, and there is no universally accepted definition. This definition applies to groups that reproduce through sexual reproduction.)

DNA: nucleic acid that carries the genetic information in all forms of life.

Gene: the basic unit of heredity, containing a sequence of DNA, at a particular location on a chromosome; codes for specific traits in an individual.

Allele: variant form of a particular gene that produce variations in inherited characteristics.

Genotype: the genetic makeup, or specific genes that an individual has.

Phenotype: the observable traits or characteristics of an individual, resulting from an interaction between its genotype and its environment.

Homozygous: having the same alleles at a particular gene locus on homologous chromosomes (e.g., RR or rr)

Heterozygous: having different alleles at a particular gene locus on homologous chromosomes (e.g., Rr).

ACKNOWLEDGEMENTS:
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ONLINE RESOURCES FOR ACTIVITY

Part 1: History of Bed Bug Control in the "History of Bed Bug Management" video located at http://njaes.rutgers.edu/bedbug/?videos#professionals.

The "History of Bed Bug Management—with Lessons from the Past" by Michael F. Potter. This article is available at http://entsoc.org/PDF/2011/AE-Potter-spring2011.pdf.

http://news.ufl.edu/2014/05/20/homemade-bedbug-trap/

The "Bed Bugs Bite Back Thanks to Evolution" article, can be found on the Understanding Evolution website at http://evolution.berkeley.edu/evolibrary/search/lessonsummary.php?&thisaudience=9-12&resource_id=418.

The "How a Leafy Folk Remedy Stopped Bed Bugs in their Tracks," article can be found at http://www.nytimes.com/2013/04/10/science/earth/how-a-leafy-folk-remedy-stopped-bedbugs-in-their-tracks.html?_r=1&.

Bed Bug Simulation Activity Instructions

Preparation and Activity Order:

1. Make copies of the bed bug genotype cards found on the next page. Cut out the cards and place them in easy-to-access sorted piles. Note: after the activity is completed, it may be helpful to store these cards in ziptop bags for future use.
2. Make copies of the student worksheets and student instructions and deliver them to the students.
3. Explain that you will be completing a simulation to see how pesticide resistance evolves in a population. Your simulation begins with an initial population of bed bugs that have been found in a hotel room. This initial population consists of **homozygous dominant** individuals (RR), **heterozygous individuals** (Rr), and one **homozygous recessive** individual (rr). **The homozygous recessive (rr) is the only genotype that will give a resistant genotype.** Note: This simulation will need to be carried out for at least five generations so a pattern can emerge.
4. Have students complete Punnett Square worksheet.
5. Read the backstory to the class.
6. Have the students create a hypothesis about the genetic cross of the two surviving bed bugs.
7. Have the students complete the simulation, record the data on the worksheet, share the data with the class, and graph the data.
8. Lead a class discussion using the "Let's Reflect and "Let's Apply" sections of the lesson.

Variations: You may wish to begin the simulation with different genotypes for your starting individuals. It is recommended to keep a resistant individual (i.e., RR x rr) but you could also start with RR x Rr or Rr x Rr (but not RR x RR, because this will never confer resistance without mutation) and see what happens. As another variation, you could use a larger starting population, but the number of offspring you select for each generation will be much larger.

Backstory to read to the class:
Your family went on a spring break trip to visit Mt. Rushmore and learn about the founders of our nation. When you arrived at the hotel, you were the first to enter the room so you could expertly check for bed bugs. It is a good thing you knew to pull back the sheets and check the mattress seams, because you found bed bug fecal spots so began to check more closely. After using your cell phone flashlight behind the headboard, you spotted several adult bed bugs, who quickly ran from the light. Your family gathered their things from the hallway, and went back down to the front desk to tell the hotel manager the news. The hotel manager thanked you for your close inspection, apologized for the inconvenience, and moved your family to a clean room. The hotel manager hired a professional pest management company to spray the room with pesticides as a control method for the bed bugs. You wanted to follow up your investigation, so asked the manager if you could come back at the end of your vacation and check the room again. You found that most of the bed bugs were killed by the pesticide application, but there were a few survivors. You collected the two survivors and had them analyzed. The two bed bugs that survived was a heterozygous individual (Rr) that was in a really good hiding spot and avoided the spray, and the one homozygous recessive individual (rr) that was in the population. The homozygous recessive individual (rr) has the genotype that confers pesticide resistance, so it was unaffected by the pesticide spray. Since these survivors were a male and female, you decided to do an experiment. What is your hypotheses about what will happen to a bed bug population that starts with these two individuals and continues for multiple generations? Write your hypothesis on your simulation worksheet. Once you have your hypothesis. Complete the data for the **starting population, 0 RR, 1 Rr, and 1 rr**.

Student Instructions for the bed bug survivor simulation.

1. You are ready to perform your genetic crosses with the bed bug survivors. These two are your starting population. **Record RR and rr as your starting population on your worksheet**.
2. These two remaining individuals in the population mate and have four offspring. You have already completed the Punnett square worksheet, so refer to that for results of genetic combinations for a Rr x rr mating. (In this case, you should have two Rr and two rr offspring.) Write down this data in your table for **generation 1.**
3. Now you would like to see what happens over multiple generations. You are looking for trends. Will the bed bugs be resistant to pesticides or non-resistant over time? Take **two Rr genotype cards** and **two rr genotype cards**, representing generation 1, and place them in a paper sandwich bag or in a folded piece of paper where the cards can be drawn without seeing the letters. These cards represent the individuals that will pair and mate to produce the next generation. For simplicity, assume that the parents of each generation do not have any more offspring and die after mating. In a natural setting, a bed bug female may mate several times and can produce up to 500 eggs in her life of 6-18 months. For this simulation, our bugs will only mate one time and will have four offspring .
4. **Draw two cards.** Take a look at your Punnett square worksheet and find a cross with those two individuals. Write down the results of that cross and record the genotype and phenotype of the four offspring on the worksheet. You are not done yet. Now take the two cards left in your bag, and add the genotype totals to the first set of offspring. The totals should be recorded as g**eneration 2**. You should have a total of eight offspring. For example, if you drew Rr x rr, you would have two offspring of Rr and two of rr for each pair. Your total would be four Rr and four rr.
5. Take eight new cards that represent generation 2 and place them in your paper bag or paper folder. In our example, you would have four Rr and four rr cards in your bag.
6. Randomly draw **two cards** from this group and record their four offspring . Do this three more times until all your cards have been drawn and record the totals as **generation 3.**
7. Take sixteen new cards that represent generation 3 and pace them in your bag or folder.
8. Randomly draw two cards from this group and record the offspring. Repeat this for each pair of cards left in your bag or folder, and record the total as **generation 4.**
9. Take new cards that represent generation 4 and place them in your bag or folder.
10. Randomly draw two cards from this group and record their four offspring as **generation 5**. Repeat this for the other card pairs. By this generation, you should have genotypes for 64 bed bug individuals. Calculate the number of non-resistant and resistant bed bugs for each generation and the total number of resistant and non-resistant bed bugs. You should have a total of 126 individuals including your two starting individuals.
11. Answer the questions on the Bed Bug Simulation Activity Worksheet.
12. Share your data with the class and graph the data.
13. Discuss the results .

Bed Bug Pesticide Resistance Simulation Activity

Before completing the simulation activity, write down what your hypothesis (what do you think will happen in the simulation)? This should be written as an *If...then...statement.* An example would be, *"If two homozygous dominant individuals are crossed, then the offspring will be non-resistant."*

My hypothesis:

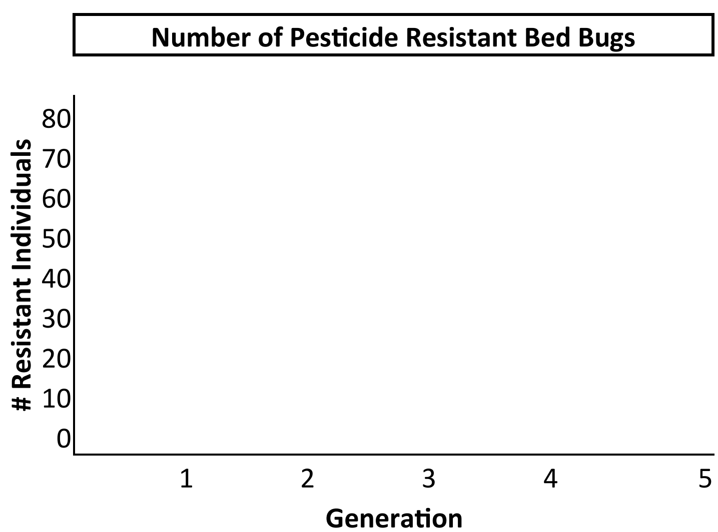
As your class completes the simulation, keep track of the data for each generation in table on the next page then answer the post activity questions and graph your data. The class data can also be combined and graphed.

Post Activity Questions:

1. What happened in the simulation? Was there pesticide resistance in the bed bug population by the fifth generation?

2. Was your hypothesis accurate? Why or why not?

3. Graph the results of the simulation. What pattern do you see (if any)?



Bed Bug Pesticide Resistance Simulation Activity Data Table. Record the genotype for each offspring then record the total number of each phenotype. Your first cross will be Rr x rr. There will be four offspring as Gen 1.

Generation	# RR	# Rr	# rr	Total of Resistant Bed Bugs	Total of Non-Resistant Bed Bugs
Starting Population (2 total)	0				
Generation 1 (4 total)					
Generation 2 (8 total)					
Draw 1					
Draw 2					
Generation 3 (16 total)					
Draw 1					
Draw 2					
Draw 3					
Draw 4					
Generation 4 (32 total)					
Draw 1					
Draw 2					
Draw 3					
Draw 4					
Draw 5					
Draw 6					
Draw 7					
Draw 8					
Generation 5 (64 total)					
Draw 1					
Draw 2					
Draw 3					
Draw 4					
Draw 5					
Draw 6					
Draw 7					
Draw 8					
Draw 9					
Draw 10					
Draw 11					
Draw 12					
Draw 13					
Draw 14					
Draw 15					
Draw 16					

Pesticide Resistance Punnett Square Worksheet

Background Information:

In this activity, the mechanism for pesticide resistance in bed bugs is genetically recessive (r). Therefore, the allele for resistance will be masked or carried in heterozygous individuals (Rr), making them non-resistant, but able to pass along the resistant allele to their offspring. Non-resistant individuals are susceptible to pesticides, so would die if exposed to them.

	Genotype	Phenotype
Homozygous Dominant	RR	Non-resistant
Heterozygous	Rr	Non-resistant
Homozygous Recessive	rr	Resistant

To determine all the possible outcomes and predict outcomes of a genetic cross, scientists use Punnett squares. Your job is to use the Punnett Squares below to figure out the **genotype** and **phenotype** of offspring produced when insects of the following genotypes mate: <u>RR x RR, Rr x Rr, RR x rr, Rr x rr, Rr x RR, and rr x rr.</u>

Each parent contributes on of their two alleles to the offspring. You work this like a multiplication problem. The alleles of each parent are put in a box on the top of side of the square. Parent one would be RR and parent two would be RR, so RxR = RR, so all the offspring would be RR. There is a 100% chance that the genotype of the offspring will be RR and the phenotype would be non-resistant in this genetic cross. This first one has been done as an example. Now, cross the other possible combinations to see if the offspring are resistant or non-resistant to pesticides.

	R	**R**
R	RR	RR
R	RR	RR

Non-resistant: 4
Resistant: 0

\# Non-resistant: ________

\# Resistant: ________

\# Non-resistant: ________

\# Resistant: ________

\# Non-resistant: ________

\# Resistant: ________

\# Non-resistant: ________

\# Resistant: ________

\# Non-resistant: ________

\# Resistant: ________

Bed Bug Pesticide Resistance Simulation Activity: Sample Answer Key

Note: Results will vary from simulation to simulation. Answers vary based upon results of the simulation. The data and answers below are the result of a single run of the simulation.

Before completing the simulation activity, write down what your hypothesis (what do you think will happen in the simulation)?

I think the resulting population will be completely (100%) resistant to pesticides, so my hypothesis would be that if a population of bed bugs contained resistant individuals, then over time, the population would become resistant.

As your class completes the simulation, keep track of the data for each generation in table below:

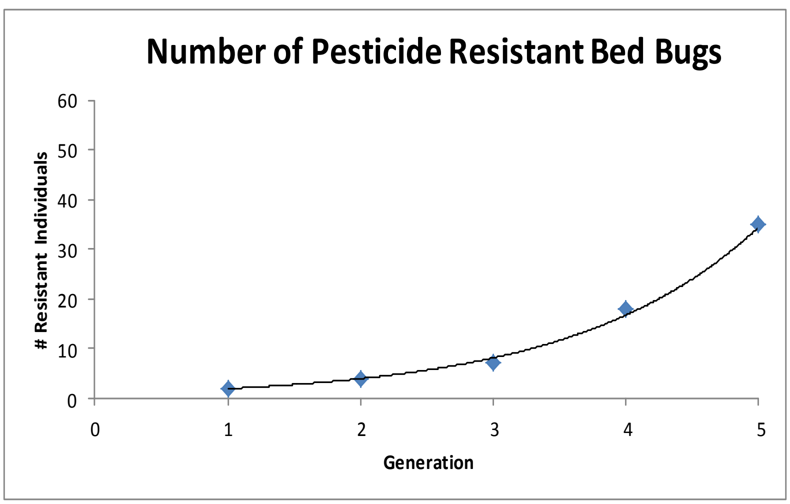
Generation	RR	Rr	rr	Total of Resistant Bed Bugs	Total of Non-Resistant Bed Bugs
Starting Population	0	1	1	1	1
Generation 1	0	2	2	2	2
Generation 2	0	4	4	4	4
Generation 3	1	6	9	7	9
Generation 4	4	14	14	18	14
Generation 5	9	26	29	35	29

Post Activity Questions:

1. What happened in the simulation? Was there pesticide resistance in the bed bug population by the fifth generation? There was pesticide resistance in just under half of the population by the fifth generation.

2. Was your hypothesis accurate? Why or why not? No, I predicted the population would be completely resistant, but just under half of the population was resistant. See "Let's Reflect" for more information about results of simulation.

3. Graph the results of the simulation. What pattern do you see (if any)? In this case, the number of resistant individuals seems to (approximately) double in each generation.



Pesticide Resistance Punnett Square Worksheet: Answer Key

Background Information:

In this activity, the mechanism for pesticide resistance in bed bugs is genetically recessive (r). Therefore, the allele for resistance will be masked or carried in heterozygous individuals (Rr), making them non-resistant, but able to pass along the resistant allele to their offspring. Non-resistant individuals are susceptible to pesticides, so would die if exposed to them.

	Genotype	Phenotype
Homozygous Dominant	RR	Non-resistant
Heterozygous	Rr	Non-resistant
Homozygous Recessive	rr	Resistant

To determine all the possible outcomes and predict outcomes of a genetic cross, scientists use Punnett squares. Your job is to use the Punnett Squares below to figure out the **genotype** and **phenotype** of offspring produced when insects of the following genotypes mate: <u>RR x RR, Rr x Rr, RR x rr, Rr x rr, Rr x RR, and rr x rr.</u>

Each parent contributes on of their two alleles to the offspring. You work this like a multiplication problem. The alleles of each parent are put in a box on the top of side of the square. Parent one would be RR and parent two would be RR, so RxR = RR, so all the offspring would be RR. There is a 100% chance that the genotype of the offspring will be RR and the phenotype would be non-resistant in this genetic cross. This first one has been done as an example. Now, cross the other possible combinations to see if the offspring are resistant or non-resistant to pesticides.

	R	**R**
R	RR	RR
R	RR	RR

Non-resistant: 4

Resistant: 0

	R	**r**
R	RR	Rr
r	Rr	rr

Non-resistant: 3

Resistant: 1

	r	**r**
R	Rr	Rr
R	Rr	Rr

Non-resistant: 4

Resistant: 0

	r	**r**
R	Rr	Rr
r	rr	rr

Non-resistant: 2

Resistant: 2

	R	**R**
R	RR	RR
r	Rr	Rr

Non-resistant: 4

Resistant: 0

	r	**r**
r	rr	rr
r	rr	rr

Non-resistant: 0

Resistant: 4

Genotype Cards for Simulation Activity

RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**

Genotype Cards for Simulation Activity

RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**
RR	**RR**	**RR**

Genotype Cards for Simulation Activity

Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**

Genotype Cards for Simulation Activity

Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**
Rr	**Rr**	**Rr**

Genotype Cards for Simulation Activity

rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**

Genotype Cards for Simulation Activity

rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**
rr	**rr**	**rr**