Herbicide Resistance through Engineered Mutation: Which Bean is a Mutant?

Enrichment Lab



Mutation in genes can be the result of genetically engineered mutations or natural mutations. This lab explores an engineered mutation for herbicide resistance and guides students to develop and conduct an experiment for determining which of 3 bean varieties carry the mutation.

Central Question:

- Does a mutation for herbicide resistance help, harm, or have no effect on bean plants?
- Which bean variety carries the EPSPS gene for glyphosate resistance?

Objectives:

- Explain that Genetically Modified Organism (GMO) technology alters the genetic material of organisms.
- Describe the basic concepts for producing glyphosate resistant GMOs.
- Develop a hypothesis and test plants for glyphosate resistance.

Related NE Science Standards:

Life Science – Heredity

- SC12.3.2 Students will describe the molecular basis of reproduction and heredity
 - SC12.3.2.c Recognize how mutations could help, harm, or have no effect on individual organism

Anticipated Length:

30 min for lab preparation (in or out of class)

20 min for experiment setup

5 min a day for 20 days in order to water and record observations

15 min on day of herbicide application

This should be when the seeds have all emerged and developed leaves, estimate around day 12.
20 min for Lab Analysis and Worksheet (in or out of class)

Lab Materials:

- 1 Variety of Glyphosate Resistant Soybean Seeds
 - Example: Roundup Ready Soybeans by Monsanto
 - Suggested Source: Local soybean farmer or co-op with beans in storage, Nebraska Soybean Board
- 2 Varieties Non Glyphosate Resistant Bean Seeds (does not need to be soybean)
 - Examples: Non-GMO Soybean, edamame beans, and other garden beans available
 - Suggested Source: Local garden center, Nebraska Soybean Board
- Glyphosate herbicide with only glyphosate
 - <u>Note</u>: some glyphosate herbicides contain additional active ingredients toxic to many plants including the glyphosate resistant plants)
 - Example: agricultural Roundup
 - Suggested Source: Local garden center or co-op, Nebraska Soybean Board
- Potting soil



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- Plant growing trays or other planters
- Masking tape for labeling trays
- Marker for labeling
- Small spray bottle for herbicide application
- Personal protective equipment for herbicide application and handling: gloves, goggles, etc.
- Space to grow plants with germination and growth conditions
- light, water, temperature ~ 77 deg. F

Disclaimer:

The use of the EPSPS gene, causing glyphosate resistance, in soybeans is proprietary and patented. Obtaining and using the soybeans shall provide NO economic gain for any party involved.

NO soybeans shall be harvested from the plants used in the laboratory.

ALL soybean seedling must be destroyed at conclusion of the lab experience.

Safety Precautions:

- Make sure that all materials are handled and used in accordance with the materials' labels and directions.
- Materials should be handled safely by appropriate persons following correct and safe lab techniques.

Teacher Notes:

This enrichment lab is designed to complement your classroom instruction on DNA and genetics. The lab is written to be either self-guided by students or enhanced with additional classroom instruction and activities. More or less teacher guidance will be required based upon your class' experience with science methods, developing experiments, creating tables, etc.

- Remove or replace packaging with labels identifying the beans. Re-label the bean varieties as A, B, and C so that students can use the letters to identify the beans, even though the beans may look different and be different varieties, this will help with forming hypothesis and creating conclusions.
- Have all materials available and space for students to store their experiments for up to 3 weeks.
- Place limitations, as needed, on materials available for each student/group experiment so that no one runs short of seeds, planters, etc.
- Provide each student with a complete copy of this lab including the Lab Information, Lab Preparation, and the Lab Analysis and Conclusion Worksheet.
- Control Group Plant and grow beans of each variety that will not be sprayed with the herbicide. This will serve as the control group for the entire class. Label each bean variety (A,B,C), and the whole group as "control." This can be delegated to students.
- Prior to glyphosate application, prepare and dilute glyphosate according to labeled directions for use and application. Train students on proper application or apply it for the students. Store and dispose according to labeled directions.
- Upon completion of student experiments, reveal the varieties of beans including the GMO for glyphosate resistance (EPSPS Gene).



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Student Name: ____



Background

A mutation, or change in the nitrogen base sequence of DNA, can be the result of genetically engineered mutations or natural mutations. Fortunately, the internal workings of cells have mechanisms in place to fix or edit any mistakes made during many natural processes like DNA replication. If DNA is altered and not corrected, either on purpose or by mistake, the process of transcription and translation will follow the DNA template and create the protein which the DNA is coded to create, thus creating a physiological and/or phenotypical change in the organisms.

A genetically engineered mutation results in a GMO, or Genetically Modified Organism. An example of a GMO is glyphosate resistant plants. Here are a few notes on glyphosate:

- a. Glyphosate is a chemical that generally kills all cells in grasses and broadleaf plants. The Roundup™ brand is an example of an herbicide with glyphosate.
- b. Some organisms NATURALLY have a gene that makes all the cells in the plant resistant to glyphosate. It is known as the EPSP Synthase (EPSPS) gene. Plants with the EPSPS gene survive exposure to glyphosate.
- c. If the EPSPS gene is added to a specific plant that does not naturally have the gene, that plant will create the proteins to resist and survive exposure to glyphosate.
- d. Beans do not naturally have the EPSPS gene for glyphosate resistance. Some GMO bean varieties have been genetically engineered and carry the gene.
- e. The use of glyphosate resistant plants allows for entire fields of crops, and the weeds included, to be exposed to glyphosate by applying a glyphosate herbicide. The chemical herbicide can kill all the plants in the field except for the plants with the EPSPS gene. This can be effective weed control by reducing weed competition for resources and therefore increasing the potential for crop growth and yield.

Procedure

- This is information for developing your experiment. This is NOT a complete lab procedure.
- Bean seeds should be planted 1 inch deep and 1 to 2 inches apart.
- Water potting soil daily, but not with a lot of water soil should be moist, not damp
- Herbicide Application:
 - Apply to all your plants on the same day once the beans have emerged and have grown true leaves, beyond cotyledons
 - Apply in accordance with label directions and teacher instructions
 - Spray in a location that is well ventilated, clear of other plants, people, and animals, outside or in a vented hood
 - Spray as a mist, rather than a stream or foam spray
 - Wear protective equipment and wash hands and arms thoroughly after spraying
- Control Group the class will plant and grow beans of each variety that will NOT be sprayed with the herbicide. These beans will serve as the control group for the entire class. Be sure to make observations of the control group along with your experiment.

Which Bean is a Mutant? — Lab Procedure



Student Name: _____

Step 1: The Lab Objective is stated below. Underline or highlight any parts of the objective that are unfamiliar or unclear to you.

Lab Objective: Design and execute a lab experiment to determine which bean variety carries the EPSPS gene for glyphosate resistance.

Step 2: Use the Background information provided and any additional resources available to you to answer these questions and understand the Lab Objective above.

1. What does glyphosate do to plants?

- 2. What does the EPSPS gene allow plants to do?
- 3. Are GMOs derived from natural or human engineered mutations?
- 4. Do beans naturally carry the EPSPS gene?
- 5. What additional questions do you have for understanding the Lab Objective? Any questions should be written here and answered before proceeding to the next step.

Which Bean is a Mutant? — Lab Procedure

IDEA BIN

Student Name: _____

Step 3: Use what you know to make predictions and construct a hypothesis. During your lab experiment you will spray glyphosate herbicide on beans with the EPSPS gene and beans without the EPSPS gene. To help write our hypothesis we will make a few predictions, identify the variables, and then write a hypothesis.

Look at the 3 bean varieties (A, B, and C) for the lab and make predictions by filling in the blanks.

My Predictions:

- a. If <u>Bean A</u> plants are sprayed with glyphosate herbicide, then the beans will (choose: survive or die) because (provide your reason)______
- b. If <u>Bean B</u> plants are sprayed with glyphosate herbicide, then the beans will (choose: survive or die) because (provide your reason)______
- c. If <u>Bean C</u> plants are sprayed with glyphosate herbicide, then the beans will (choose: survive or die) because (provide your reason)______

Identify the variables in your predictions and lab experiment:

- List the 3 Independent Variables: ______
- List the Controlled Variable:

Write a hypothesis:

A good format to follow is:

If (We do this to the test subject with the Independent Variable), then (This will happen to the test subject in relation to the Dependent Variable) because (Support the statement with information you know).

Your Hypothesis:

Which Bean is a Mutant? — Lab Procedure

Student Name: _____



Step 4: Design your experimental protocol. Outline a lab experiment to determine which of the 3 bean varieties carry the EPSPS gene for glyphosate resistance. Provide enough information so that someone else could use your outline to conduct the same experiment.

Your outline should include:

- What beans to plant
- How many
- How to plant
- How to label plants/planters
- How to care for the beans
- What to spray the plants with
- When and how to spray the plants
- Other steps for successful completion of the experiment.
- Data charts to gather observations (with Qualitative and Quantitative observations)

Use additional paper for outline.

Step 5: Upon teacher approval, setup and start your lab experiment.

Step 6: Collect data in your table or tables of observations, data, and activities (planting, watering, spraying, etc.). You will make observations of your experiment as well as the control group. Make lab observations and notes daily.

Use additional paper for table(s).

Lab Analysis and Conclusion Worksheet

Complete this section after completing of your lab

IDEA BIN

Student Name: __

- 1. What were you trying to find out with your experiment?
- 2. What, if any, changes would you make in your experiment, setup and care, or observations you recorded? Why?
- 3. Was your hypothesis correct or incorrect? Explain why?
- 4. Predictions:
 - Was your prediction for Bean A correct or incorrect? Tell why.
 - Was your prediction for Bean B correct or incorrect? Tell Why.
 - Was your prediction for Bean C correct or incorrect? Tell Why.
- 5. Which bean variety carries the EPSPS gene for glyphosate resistance?
- 6. Does a mutation for glyphosate resistance seem to help, harm, or have no effect on a plant? Explain.
- 7. Why would food and crop plants with the EPSPS gene be developed and used?
- 8. How did this lab help you understand engineered mutations and GMOs?
- 9. What additional experiment would you like to do in order to answer a related question?