Dye Electrophoresis Lab

Mendel's Peas

miniPCR bio Learning Lab™
Dye Electrophoresis Lab:
Mendel's Peas

Version: 1.0
Release: July 2023

Contents

At a glance

This miniPCR bio Learning Lab™ was created to give beginner students hands-on experience with biotechnology. The use of safe and affordable dyes to simulate DNA samples makes it easier than ever to bring gel electrophoresis to your classroom!

Lab overview

Some of Mendel's peas have been discovered in an old notebook! Can you confirm which gene Mendel was studying in the 1800s?

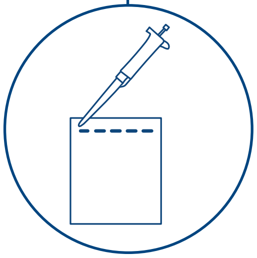
This lab connects traditional Mendelian genetics with our modern understanding of DNA and inheritance. Students play the role of plant geneticists testing peas from Mendel's notebook. Gel electrophoresis is used to analyze simulated DNA samples from Mendel's peas to examine how genetic differences control pea shape.

TECHNIQUES	TOPICS	LEVEL	WHAT YOU NEED
Micropipetting	Genotype to phenotype	Middle school	Micropipette
Gel electrophoresis	Mendelian inheritance	General high school	Gel electrophoresis system
	Biotechnology		

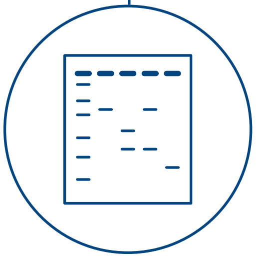
Planning your time

SINGLE CLASS PERIOD: 45 MINUTES if gels are prepared in advance

Gel electrophoresis



Interpret results



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Additional supports

At miniPCR bio™, we are committed to preparing students to be successful in the laboratory through high quality curriculum and training. We have created an extensive set of resources to help your students succeed in molecular biology techniques, all of which are available for free at https://www.minipcr.com/tutorials/.

The resources most relevant to this lab are listed below.

Micropipetting: Video, worksheet, and hands-on activities to train students in the basic use of a micropipette.

Gel electrophoresis: Video and worksheet activity instructing students on the fundamentals and practice of agarose gel electrophoresis.

PCR: While students do not perform PCR in this lab, the samples they analyze represent PCR products. If you want to discuss PCR in more detail with your students, we have a video and worksheet activity instructing students on the fundamentals and practice of PCR.

For answers to the lab study questions, email answers@minipcr.com. Please include the name of the lab, as well as your name, school, and title in the body of the email.

Materials needed

Supplied in kit (KT-1403-01)

- Materials are sufficient for 8 lab groups.
- Gel reagents can be stored at room temperature.
- Refrigeration is recommended for blue dye samples to prevent evaporation.
- Reagents must be used within 12 months of shipment.

Reagents and supplies	Amount provided in kit	Amount needed per lab group	Storage
Blue dye samples	**8 Load Ready™ Strips**	**1 Load Ready™ Strip**	Refrigerator
Agarose Tabs™	**8 Tabs**	**1 Tab** per gel (if using a Bandit™ or blueGel™ electrophoresis system)	Room temp.
TBE buffer	**Supplied as powder** Sufficient to prepare 600 ml	**60 ml** per gel (if using a Bandit™ or blueGel™ electrophoresis system)	Room temp.

Required equipment

Available at minipcr.com

Item	Quantity needed
Horizontal gel electrophoresis system *e.g.*, Bandit™ STEM Electrophoresis Kit or blueGel™ electrophoresis system *Note: This lab is compatible with any horizontal gel electrophoresis system	**Each group needs 6 wells in an agarose gel** Groups can share gels if your gel electrophoresis system is large enough
Micropipettes and tips 2-20 µl adjustable volume or 10 µl fixed volume	**1 pipette per group**

Other materials supplied by user

- Microwave or hot plate to dissolve agarose
- Distilled water for making agarose gels and dissolving TBE buffer
- Screw top bottle to store prepared TBE buffer
- Heat-resistant flask or beaker to dissolve agarose
- Disposable laboratory gloves
- Protective eye wear
- Permanent marker
- Cup to dispose of tips

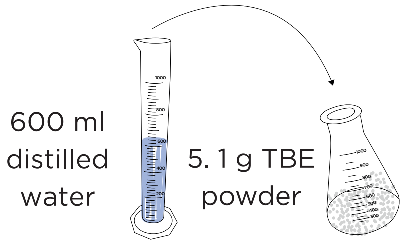
Lab setup

- The following activities can be carried out by the instructor ahead of class.
- Reagents are sufficient to be used with 8 student groups.
- Reagents are stable at room temperature.

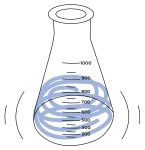
Gloves and protective eyewear should be worn for the entirety of this lab.

A. Prepare TBE buffer

1. **Combine TBE powder and 600 ml distilled water.**
    - Obtain a heat resistant container such as a glass Erlenmeyer flask or beaker that is at least 1 L in volume.
    - The lab kit comes with a pouch of TBE powder. Empty entire container of TBE powder (5.1 g) into the flask or beaker.
    - Add 600 ml distilled water.



2. **Dissolve TBE powder.**
    - Stir or intermittently shake solution until TBE powder is dissolved (this may take up to 10-15 minutes).
    - You may warm as necessary to help dissolve the powder.
    - It is normal for a small amount of powder to remain undissolved after 15 minutes. Small amounts of undissolved powder will not affect performance.



3. **Store prepared TBE buffer.**
    - TBE buffer can be stored in an airtight container at room temperature for at least three months.
    - Discard unused TBE buffer if it becomes cloudy.

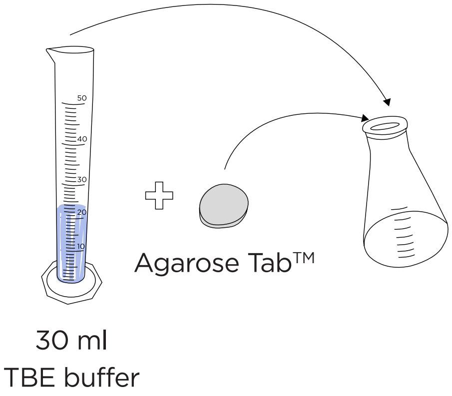
B. Make gels

Important Gel Pouring Notes

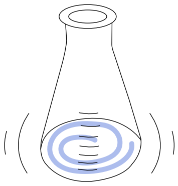
- Gels can be prepared up to five days ahead of time. Store in an airtight container at room temperature.
- These instructions are designed for use with the pre-weighed Agarose Tabs™ provided in the lab kit.
- One Agarose Tab™ will yield one gel for use in either a Bandit™ or blueGel™ electrophoresis system by miniPCR bio™.
- If using a different electrophoresis system, these instructions may need to be adjusted according to the manufacturer's instructions. Each tab contains 0.5 g of agarose, and you can use gels of any percentage between 1-2%.

See detailed assembly and gel pouring instructions for the Bandit™ STEM Electrophoresis Kit https://www.minipcr.com/bandit-assembly/

1. **Prepare an agarose solution.**
    - Obtain a heat-resistant container such as a glass Erlenmeyer flask or beaker that holds at least three times the volume you wish to add.
    - Combine 30 ml room temperature TBE buffer and one Agarose Tab™ for each gel you plan use in a Bandit™ or blueGel™ electrophoresis system.
    - Allow the tabs to soak until they fully disintegrate (this might take a few minutes).
    - Swirl the flask or beaker to ensure the tabs have fully disintegrated before heating.



2. **Heat solution.**
    - Expect to heat for about 60 seconds per 30 ml of liquid in a standard microwave.
    - Heat until the solution boils and continue until agarose is fully dissolved. No agarose particles should remain.



Caution: The solution may boil over the top of some containers. The solution will be very hot.

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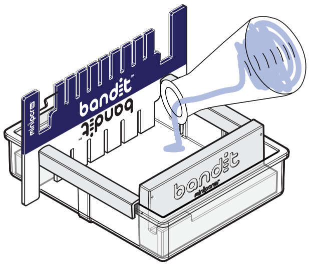
3. **Set up your gel casting system.**
    - You will need six lanes per gel.
    - If using a Bandit™ STEM Electrophoresis Kit:
        - Make sure Electrodams™ are firmly in place before pouring gel.
        - Place the comb approximately 1 cm from the black Electrodam™.

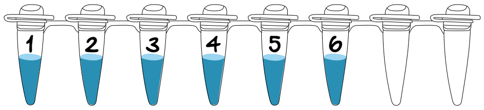
4. **Pour the agarose solution into the prepared casting setup.**
    - The agarose solution should cover the bottom of the gel tray and the bottom 3 mm of the comb (roughly the bottom 1/3 of the comb).
    - Note: Because this lab uses colored dyes as experimental samples, there is no need to add DNA stain.



5. **Allow gel to solidify completely.**
    - Gel is ready when cool and firm to the touch, which typically takes about 10 minutes.
    - Gels can be stored in an airtight container at room temperature for five days before use.
    - You can remove the comb and disassemble the gel casting apparatus before storing the gel.

C. Label Load Ready™ Strips

- On each Load Ready™ Strip, label the tubes containing blue dye 1-6. There should be two empty tubes after tube 6.
- Alternatively, have students label the tubes when they receive their Load Ready™ Strip.
- Note: If Load Ready™ Strips were stored at room temperature instead of in the refrigerator, evaporation may occur. Each tube should have at least 20 µl of dye. If significant evaporation has occurred, dye samples may become viscous and can be difficult to pipette. If this is the case, you can add 10 µl of distilled water to each sample.



D. Distribute supplies and reagents to lab groups

CHECK	At the start of this experiment, every lab group should have:	Amount
	Load Ready™ Strip containing blue dye samples	1
	2-20 µl micropipette or 10 µl fixed volume micropipette	1
	Micropipette tips	At least 6
	Six wells in an electrophoresis gel	

Reducing plastic waste

To reduce plastic waste, you may instruct your students not to change pipette tips between samples. Reusing tips will not affect the results of this lab. While best practices generally dictate that pipette tips should always be changed between samples, we also believe it is important to reduce waste when possible, and encourage you to take that into consideration in your instruction.

Student's Guide

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Background information

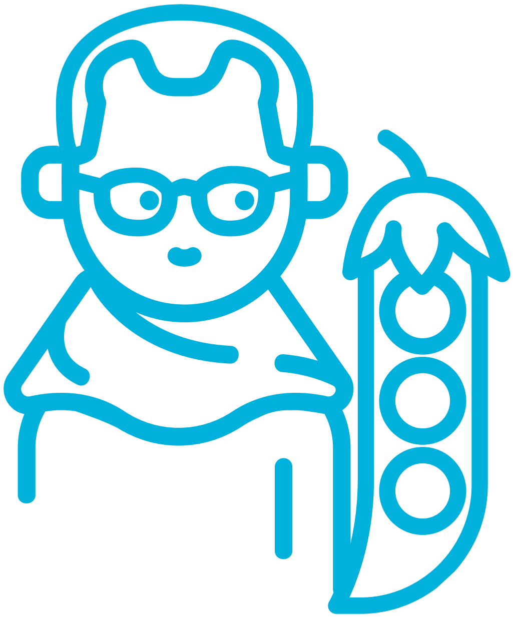
A pioneering geneticist: Gregor Mendel

1

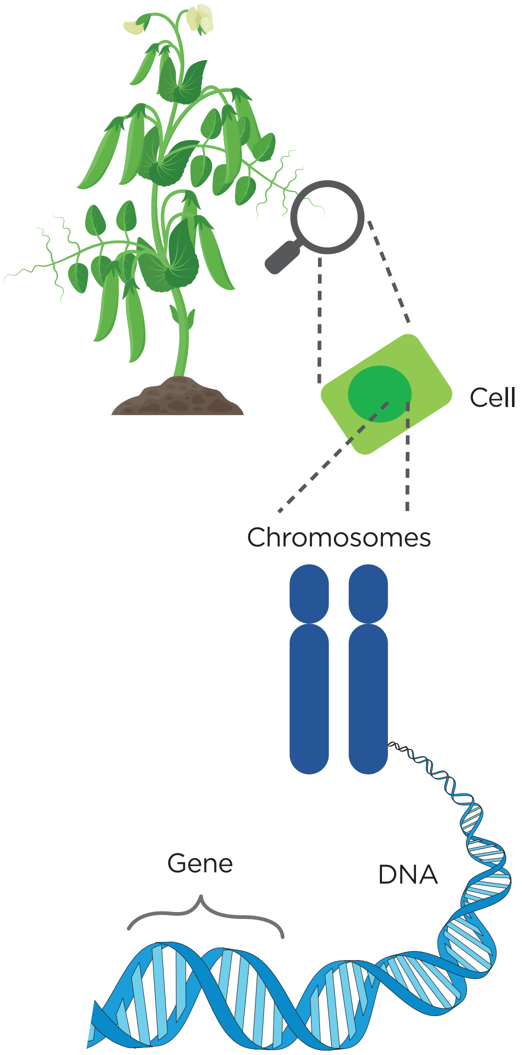
- Many traits are **inherited**, meaning they are passed from parent to offspring.
- In the 1800s, Gregor Mendel figured out basic patterns of inheritance by breeding pea plants and observing whether the offspring looked like either parent plant.
- Mendel studied inherited traits that had two forms in pea plants. For example, peas can either be round or wrinkled.

2

- Through his experiments, Mendel came to believe that pea plants receive something from each parent that defines the plant's appearance.
- He did not know what this material was, but he symbolized it using capital and lowercase letters like A and a.
- Your goal in today's lab is to connect Mendel's A and a with our modern understanding of inheritance.



DNA and genes



1

- Today, we know that all organisms inherit traits from their parents through DNA.
- **DNA** contains instructions for the cell and is found in structures called **chromosomes**.
- The cells of many organisms, including humans and pea plants, have two copies of each chromosome, one inherited from each parent.

2

- A **gene** is a section of DNA that contains a specific instruction for the cell.
- The instructions in an organism's genes determine its inherited traits.
- For example, pea plants have a gene that contains instructions that determine the shape of the pea.

3

- Each gene can come in different versions. We call different versions of the same gene **alleles**.
- Alleles of the same gene carry different instructions.
- For example, there are two alleles for the gene that determines pea shape. One allele has instructions that lead to round peas, and the other allele has instructions that lead to wrinkled peas.

4

- Even though Mendel performed his experiments before we knew that DNA underlies inheritance, his work showed how alleles pass from parents to offspring.
- Today, we know that different alleles of the same gene carry different instructions because they have differences in their DNA.

Background: Stop and think

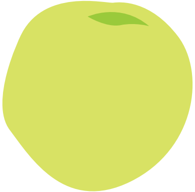
Circle the words that complete the sentence:

Q1. (Alleles/DNA molecules) are different versions of the same (gene/chromosome).

Mendelian inheritance

1

- Today, we know that Mendel was tracking alleles in his pea breeding experiments.
- Mendel coined the terms dominant and recessive to explain how some traits can skip a generation. Now we use the same language to describe the relationship between alleles of the same gene.
- A single copy of a **dominant** allele of a gene will produce the dominant trait.
- A **recessive** trait will only be present if both alleles are recessive.



Round peas
Dominant trait
AA or Aa

Wrinkled peas
Recessive trait
aa

2

- Mendel showed that having round peas is dominant and having wrinkled peas is recessive. This means that peas can only be wrinkled with two copies of the recessive allele.
- Scientists today represent alleles the same way Mendel did: an uppercase letter symbolizes the dominant allele, and a lowercase letter symbolizes the recessive allele.

Background: Stop and think

Circle the word(s) that complete each sentence:

Q2. Mendel showed that having (round/wrinkled) peas is dominant. He used capital and lowercase letters to track the inheritance of the pea shape trait. Mendel showed that a pea that was Aa would have (round/wrinkled) peas.

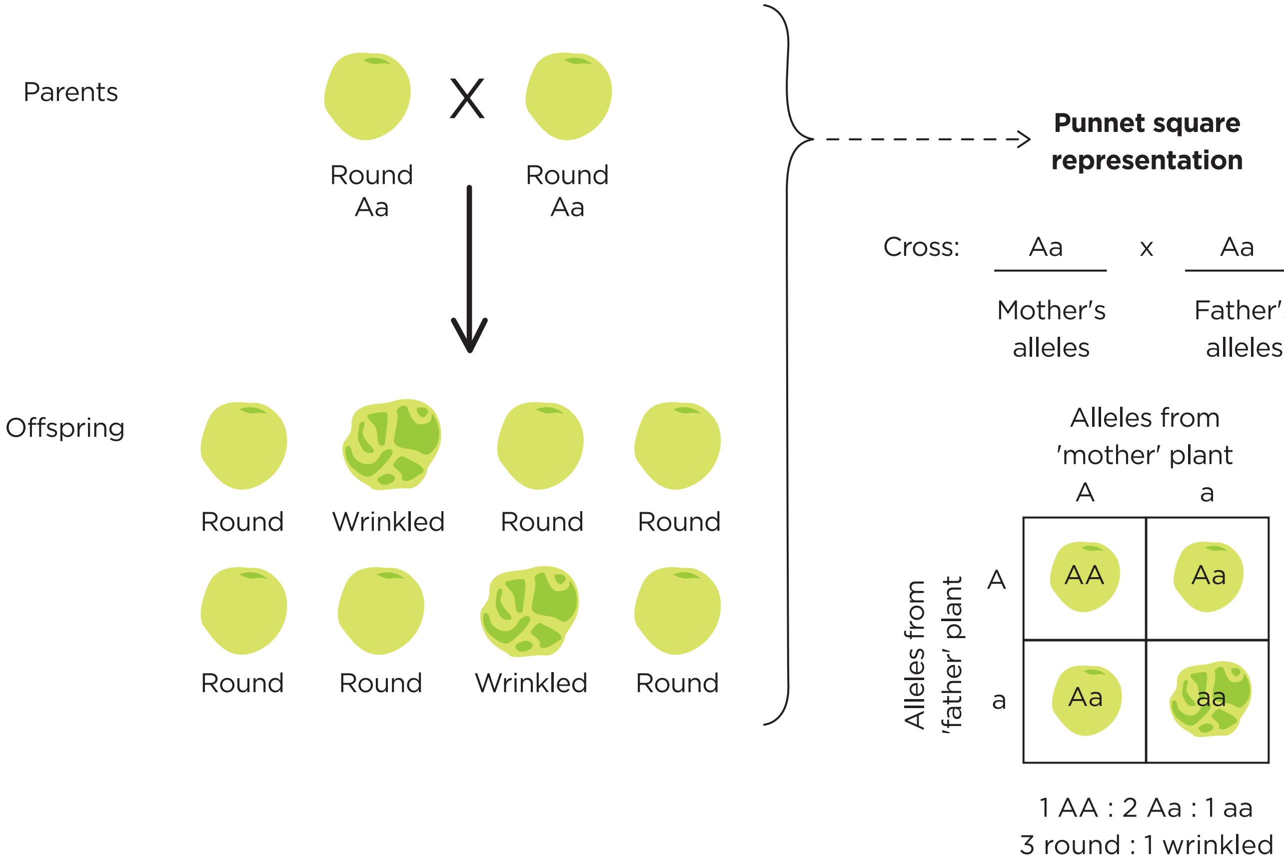
Genetics is complicated!

- In this lab, we are studying simple **Mendelian inheritance** where a single gene with a dominant allele and a recessive allele determines pea shape.
- Examining a simple trait like pea shape serves as a good entryway to inheritance, but it is important to keep in mind that most heritable traits are influenced by many genes and/or environmental factors. For example, human height is influenced by many genes, but it is also highly dependent on environmental factors such as diet.

Punnett squares

1

- Alleles are often shown in the context of a **Punnett square**, a diagram that shows the possible combinations of alleles in offspring that could be born to a pair of parents.
- Punnett squares are a useful tool to see how traits are passed from parents to offspring.



2

- Like Mendel, Punnett squares use letters to represent alleles of a gene.
- In this lab, you will connect Mendel's A and a with our modern understanding of DNA and alleles!

Background: Stop and think

Q3. Mendel studied seven different traits in pea plants. He also showed that having purple flowers is dominant and having white flowers is recessive. Use this information to answer the following questions.

- Use the letter B to represent dominant purple flower allele.
- Use the letter b to represent the recessive white flower allele.

Cross: **Bb** x **bb**

Mother's alleles

Father's alleles

A. You have a mother plant that is Bb. This plant will have (purple/white) flowers.

B. You have a father plant that is bb. This plant will have (purple/white) flowers.

C. Use the Punnett square to the right to predict the result of mating these two plants.

D. What are the expected allele ratios for the offspring plants?

__ BB : __ Bb : __ bb

E. What ratios do you expect for the offspring peas' appearance?

__ Purple : __ White

Alleles from 'mother' plant

Alleles from 'father' plant

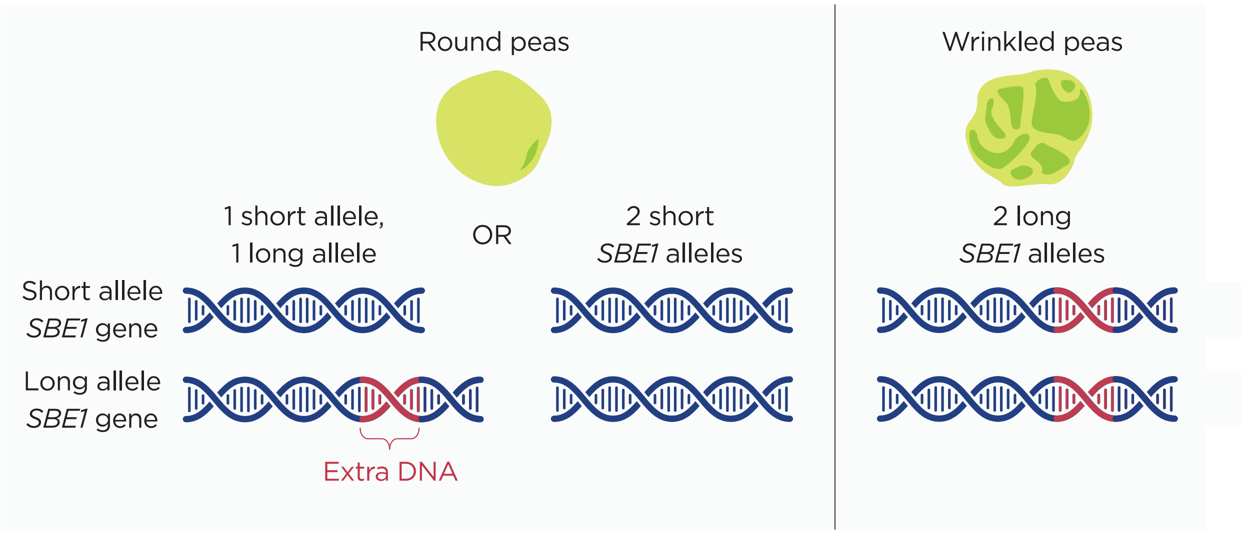
The search for Mendel's genes!

1

- We have known the inheritance pattern for pea seed shape since Mendel's experiment: round is dominant and wrinkled is recessive.
- But even with modern tools, linking inherited traits to specific genes can be tricky. In fact, scientists are still trying to figure out exactly which genes Mendel was studying!

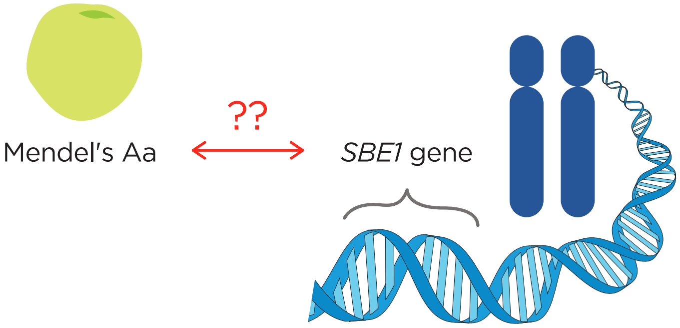
2

- You are a plant geneticist, and your lab discovered two alleles of a gene called ***SBE1*** that controls pea shape.
- The *SBE1* alleles are the same, except one has extra DNA near the end of the gene. We will refer to them as the short allele and the long allele.
- Your team has shown that the short *SBE1* allele is dominant and leads to round seeds.



3

- You suspect that the short and long alleles of the *SBE1* gene might be the A and a alleles that Mendel was tracking...
- But without access to the actual peas Mendel was using in the 1800s, there is no way to know for sure.



Background: Stop and think

Q4. Indicate which *SBE1* allele each description corresponds to:

- A. Two copies of this allele yields wrinkled peas
- B. Dominant *SBE1* allele
- C. This allele has extra DNA compared to the other allele
- D. Recessive *SBE1* allele

- I. Short *SBE1* allele
- II. Long *SBE1* allele

Today's lab

Historians have just discovered one of Mendel's lost notebooks. Included in Mendel's notes are six dried peas. You can now test your hypothesis that the short and long alleles of the *SBE1* gene correspond to the A and a alleles that Mendel was tracking!

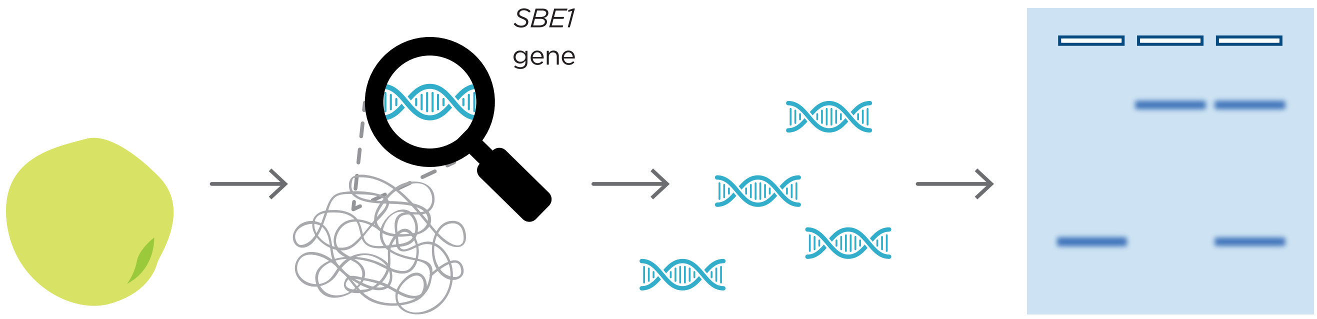
To accomplish this you will:

- **Determine which *SBE1* alleles are present in the peas from Mendel's notebook**
- **Compare each pea's *SBE1* alleles with Mendel's notes**

The ink in Mendel's notebook was smeared and you can't make out his notes for some of the peas

DNA testing

To study the peas' *SBE1* alleles, the samples will be tested as follows:



Step 1:
DNA collection

Step 2:
PCR

Step 3:
Gel electrophoresis

Already completed

You'll do this step

1

- First, DNA is taken out of the pea cells. Only a tiny piece of pea is needed.
- Someone else in your research lab already performed this step.

2

- Next, a process called **PCR** is used to copy just the *SBE1* gene from the pea's DNA.
- Someone else already performed this step too.

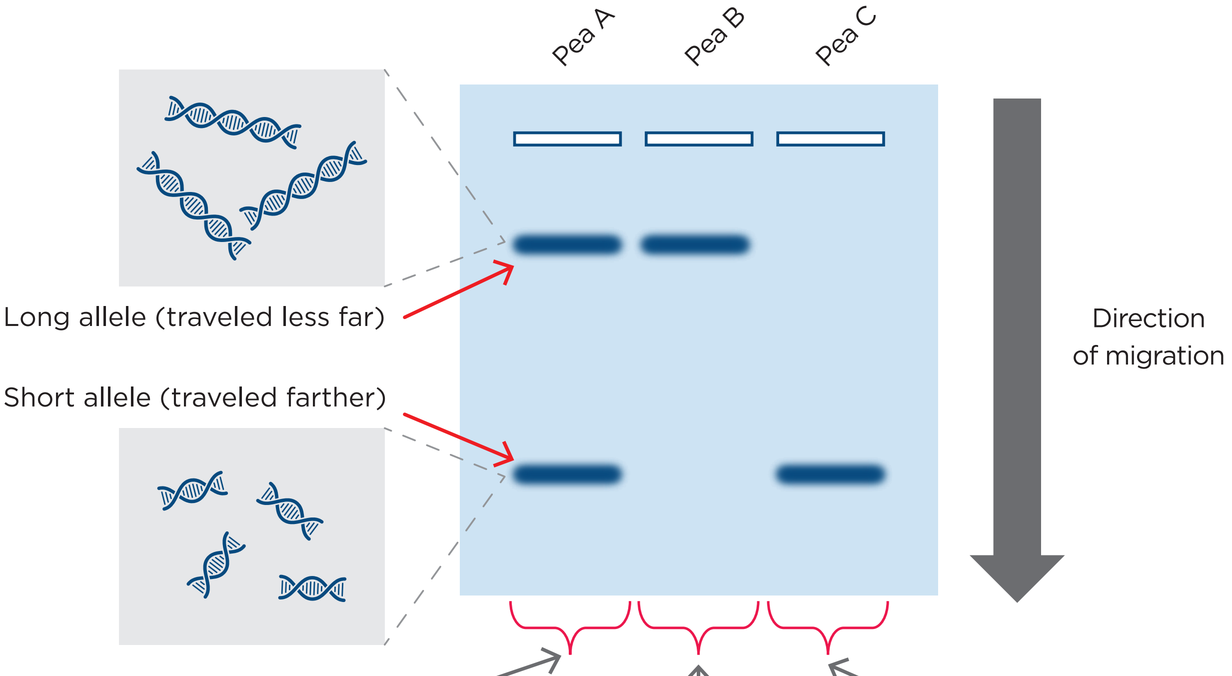
3

- Your assignment is to perform the last step of the DNA testing: gel electrophoresis.
- This will allow you to determine which *SBE1* alleles each pea has.

Interpreting gel electrophoresis results

1

- You will use gel electrophoresis to study the *SBE1* gene.
- **Gel electrophoresis** separates pieces of DNA by size. Smaller pieces of DNA travel farther through an **agarose gel**.
- Because the alleles of the *SBE1* gene differ in length, you can use gel electrophoresis to identify which *SBE1* alleles each of Mendel's peas has.
- Let's walk through gel electrophoresis results for three hypothetical peas to practice interpreting *SBE1* gel results.



2

- Pea A has two bands. This tells us that the *SBE1* DNA was two sizes.
- Because Pea A has both bands, we know it has one copy of the long *SBE1* allele and one copy of the short *SBE1* allele.

3

- Pea B only has one band. This tells us that all of the *SBE1* DNA was the same size.
- Because the DNA traveled less far, we know Pea B has two copies of the long *SBE1* allele.

4

- Pea C only has one band. This tells us that all of the *SBE1* DNA was the same size.
- Because the DNA traveled farther, we know Pea C has two copies of the short *SBE1* allele.

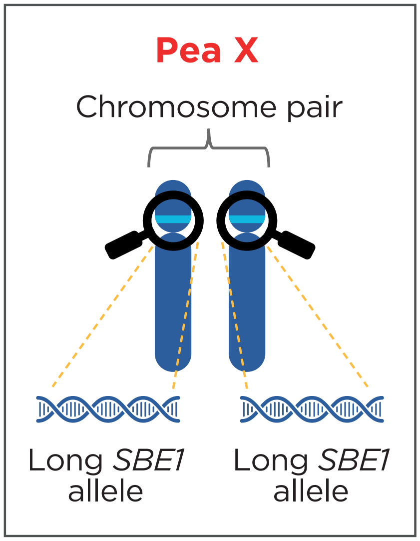
Background: Stop and think

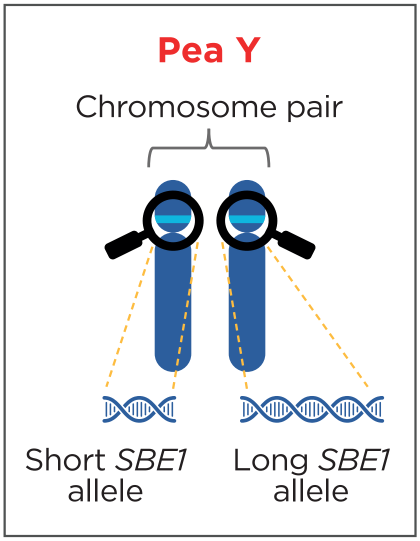
Q5. Why is gel electrophoresis a good tool for studying the different alleles of the *SBE1* gene in peas?

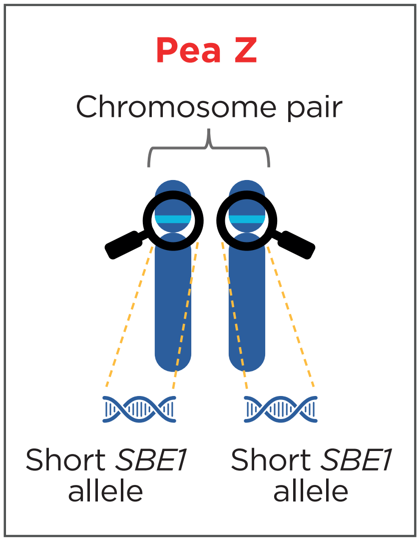
A. Because it allows you to make copies of the *SBE1* gene to study
B. Because peas have two copies of each chromosome
C. Because it allows you to extract DNA from cells
D. Because the *SBE1* alleles are different lengths

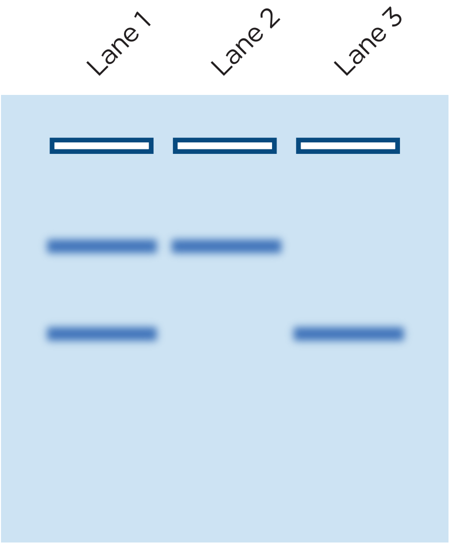
Q6. Match the potential allele combinations shown below for Peas X, Y, and Z with the corresponding gel results shown in lanes 1, 2, or 3.

A. Pea X	1. Lane 1
B. Pea Y	2. Lane 2
C. Pea Z	3. Lane 3









Glossary

Inherited: A trait is inherited when it is passed from parents to offspring through DNA .

DNA: DNA contains the instructions for the cell and is passed down from parent to offspring.

Chromosome: The structures that store DNA in the cell. Organisms like humans and pea plants have pairs of chromosomes, with one copy of each chromosome inherited from each biological parent.

Gene: A region of DNA that contains a single set of instructions. Different genes correspond to different traits.

Allele: One of two or more alternative versions of the same gene. Different alleles of the same gene have differences in the DNA.

> **Dominant:** Some alleles have a relationship called dominant/recessive. A single copy of a dominant allele of a gene will produce the corresponding dominant trait.
>
> **Recessive:** Some alleles have a relationship called dominant/recessive. A recessive trait will only be present if both alleles of a gene are recessive.

Mendelian inheritance: The inheritance pattern described by Mendel in his study of pea plants. Mendel tracked traits that are determined by a single gene with two alleles. The alleles have a fully dominant/recessive relationship.

Punnett square: A diagram that shows allele combinations in offspring that could be born to a set of parents.

***SBE1* gene:** A gene that controls whether peas are round or wrinkled. Scientists suspect this was the gene that Mendel was tracking in his experiments.

Polymerase chain reaction (PCR): A method used to make many copies of a DNA segment you are interested in studying. For more detailed information on PCR, refer to https://www.minipcr.com/polymerase-chain-reaction/.

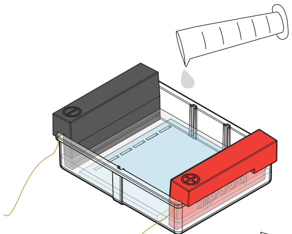
Gel electrophoresis: A method that separates pieces of DNA by length. For more detailed information on electrophoresis, refer to https://www.minipcr.com/gel-electrophoresis/.

Agarose gel: A type of gel commonly used for gel electrophoresis. Agarose is a sugar that comes from seaweed. At the microscopic level, the inside of an agarose gel looks like a web or a sponge. Small molecules can move through the holes with relative ease, but larger molecules get slowed down. This allows you to separate molecules of different sizes.

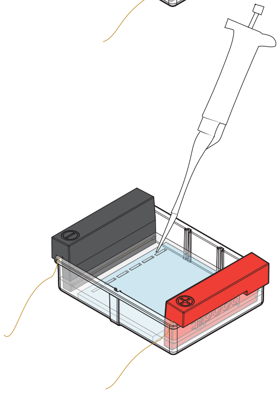
Laboratory guide

! Protective gloves and eyewear should be worn for the entirety of this experiment.

See detailed assembly and gel pouring instructions for the Bandit™ STEM Electrophoresis Kit https://www.minipcr.com/bandit-assembly/



1. **Submerge your gel in enough TBE buffer to just cover the gel and fill the wells.**
    - If using a Bandit™ or blueGel™ electrophoresis system you will need approximately 30 ml of TBE buffer.
2. **Use a micropipette to load samples onto the gel from the corresponding tubes in your Load Ready™ Strip.**
    - Lane 1: 10 µl Pea 1 DNA
    - Lane 2: 10 µl Pea 2 DNA
    - Lane 3: 10 µl Pea 3 DNA
    - Lane 4: 10 µl Pea 4 DNA
    - Lane 5: 10 µl Pea 5 DNA
    - Lane 6: 10 µl Pea 6 DNA

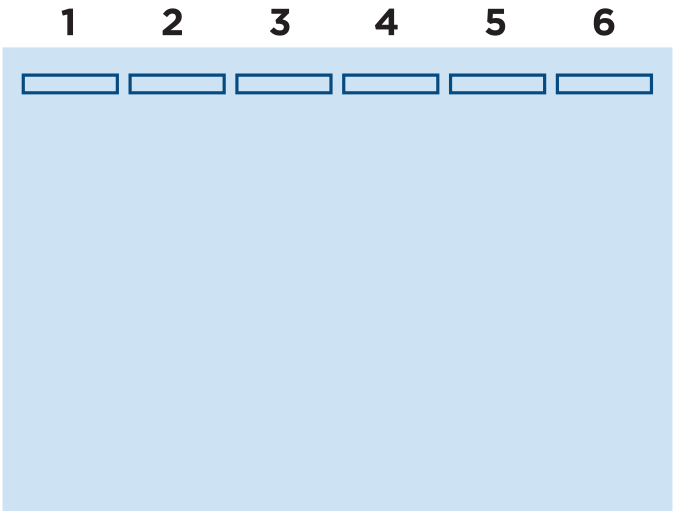


3. **Connect the electrodes and turn on your gel electrophoresis system.**
4. **Run the gel for 15-20 minutes or until there is sufficient separation between the bands.**
    - Times are based on Bandit™ and blueGel™ electrophoresis systems. If using other gel electrophoresis systems, separation time may vary.
    - Longer electrophoresis times will result in better separation.
    - Placing the gel over a white background will make it easier to see your results.

Pre-lab study questions

Critical thinking

Pea	Appearance	Mendel's notes
1	Round	Aa
2	Round	AA
3	Wrinkled	aa
4	Round	??
5	Round	??
6	Wrinkled	??



1. You know the alleles for Peas 1-3 from Mendel's notes. On the gel above, draw what your gel would look like for Peas 1-3 if your experimental results support your hypothesis that the short and long *SBE1* alleles were the A and a alleles Mendel was tracking. Be sure to label the short and long *SBE1* alleles.

2. You can't read Mendel's notes on the alleles for Peas 4-6, but you can make some predictions about which alleles they have based on what you know about the inheritance of pea shape. Abbreviate the alleles as A and a.
   A. Round peas: Peas 4 and 5

   Possible alleles:
   Explain your reasoning:

   B. Wrinkled peas: Pea 6

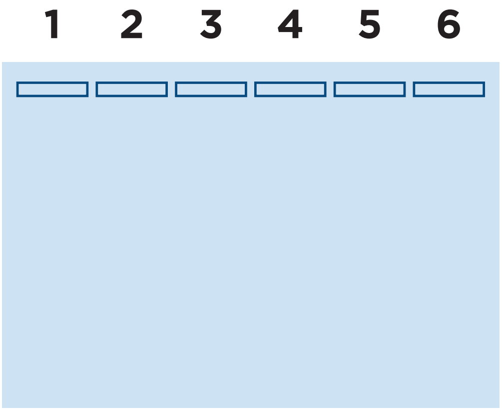
   Possible alleles:
   Explain your reasoning:

3. Use your answers from question 2 to draw what your gel would look like for Peas 4-6 if your experimental results support your hypothesis. Use a dashed line to draw alleles that you cannot predict with certainty based on the pea's appearance.

Post-lab study questions

Interpreting results

1. Use the image of a gel on the right to draw what your gel looks like. For each sample, draw the bands that you see on your actual gel.



2. Label each band as either the short allele or the long allele of the *SBE1* gene.

3. Record your results in the table below. The first row has been filled out for you as an example.

Pea	**Appearance**	**Mendel's notes**	***SBE1* gel results**
1	Round	Aa	Long allele, short allele
2	Round	AA	
3	Wrinkled	aa	
4	Round	??	
5	Round	??	
6	Wrinkled	??	

Critical thinking

Compare the results in the “Mendel's notes” column and the “*SBE1* gel results” column to answer the following questions.

4. Do your experimental results support your hypothesis that the short and long *SBE1* alleles correspond to the A and a alleles Mendel was tracking? Explain your reasoning.

5. Assuming your hypothesis is correct, what would Mendel’s notes for Peas 4, 5, and 6 have read before the ink was smeared? Explain your reasoning.

CER table

Fill in the table based on your results from the lab. Use the rubric on the next page to help your answers.

Question:

Do your experimental results confirm your hypothesis that the short and long SBE1 alleles correspond to the A and a alleles that Mendel was tracking?

Claim

Make a clear statement that answers the above question.

Evidence

Provide data from the lab that supports your claim.

Reasoning

Explain clearly why the data you presented supports your claim. Include the underlying scientific principles that link your evidence to your claim.

Score	4	3	2	1
CLAIM A statement that answers the original question/problem.	Makes a clear, accurate, and complete claim.	Makes an accurate and complete claim.	Makes an accurate but incomplete or vague claim.	Makes a claim that is inaccurate.
EVIDENCE Data from the experiment that supports the claim. Data must be relevant and sufficient to support the claim.	All of the evidence presented is hightly relevant and clearly sufficient to support the claim.	Provides evidence that is relevant and sufficient to support the claim.	Provides relevant but insufficient evidence to support the claim. May include some non-relevant evidence.	Only provides evidence that does not support claim.
REASONING Explain why your evidence supports your claim. This must include scientific principles/ knowledge that you have about the topic to show why the data counts as evidence.	Provides reasoning that clearly links the evidence to the claim. Relevant scientific principles are well integrated in the reasoning.	Provides reasoning that links the evidence to the claim. Relevant scientific principles are discussed.	Provides reasoning that links the evidence to the claim, but does not include relevant scientific principles or uses them incorrectly.	Provides reasoning that does not link the evidence to the claim. Does not include relevant scientific principles or uses them incorrectly.

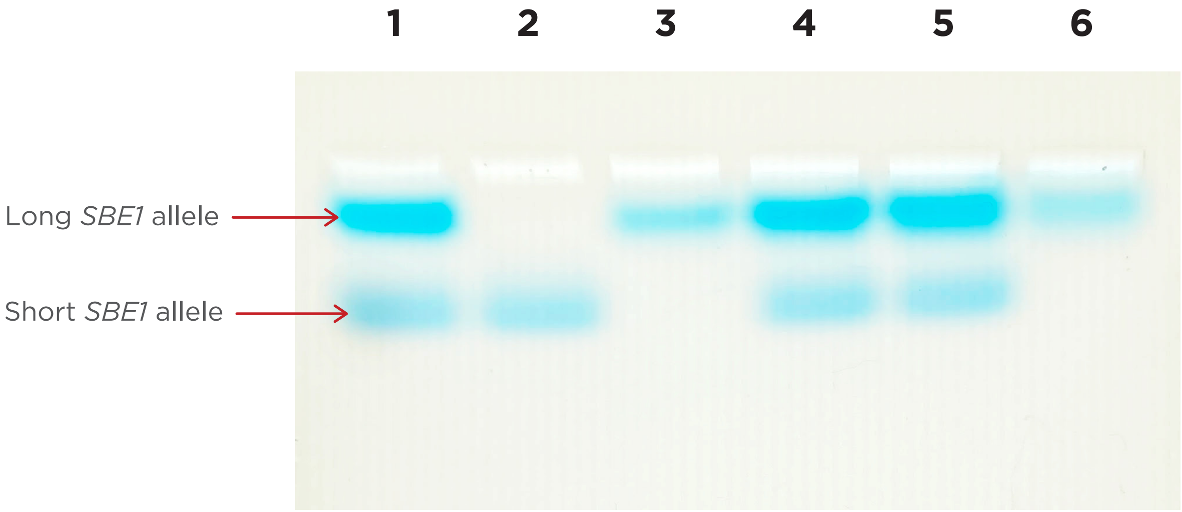
We recommend that teachers use the following scale when assessing this assignment using the rubric. Teachers should feel free to adjust this scale to their expectations.

Rubric score	3	4	5	6	7	8	9	10	11	12
Equivalent	55	60	65	70	75	80	85	90	95	100

Instructor’s Guide

Expected results

Gel electrophoresis results are expected to resemble the image below.



This image represents results obtained after a 20 minute run using the Bandit™ STEM Electrophoresis Kit.

Interpretation:

- Peas 1, 4, and 5 have one copy of the long *SBE1* allele and one copy of the short *SBE1* allele.
- Pea 2 has two copies of the short *SBE1* allele.
- Peas 3 and 6 have two copies of the long *SBE1* allele.

The short *SBE1* allele matches Mendel's A allele and the long *SBE1* allele matches Mendel's a allele.

Pea	Appearance	Alleles	*SBE1* gel results
1	Round	Aa	Long allele, short allele
2	Round	AA	Short allele, short allele
3	Wrinkled	aa	Long allele, long allele
4	Round	(AA or Aa)	Long allele, short allele
5	Round	(AA or Aa)	Long allele, short allele
6	Wrinkled	(aa)	Long allele, long allele

For answers to the lab study questions, email answers@minipcr.com. Please include the name of the lab, as well as your name, school, and title in the body of the email.

Notes on lab design

This lab serves as an introduction to the relationship between genotype and phenotype. We believe our approach provides the right balance between intellectual engagement, inquiry, and accessibility. The design of this lab has simplified certain elements to achieve these goals.

- We omit some common genetics vocabulary, including the terms phenotype, genotype, homozygous, and heterozygous. Teachers are encouraged to use these terms if appropriate for the level of your class.
- We use negatively charged dyes to simulate DNA during gel electrophoresis. This allows for the samples to be directly visualized in the gel without the need for additional staining.
- This lab does not include a detailed explanation of the science behind gel electrophoresis or PCR. However, miniPCR bio™ has published a library of resources that can be used to that effect (refer to https://www.minipcr.com/tutorials/). Bandit™ users can also take advantage of From Circuits to Molecules (https://links.minipcr.com/circuitsTG), an educational activity that walks students through the process of building their Bandit™ system to better understand how gel electrophoresis works.
- The scenario that historical samples of Mendel's peas have been rediscovered is fictional, but research does suggest that the *SBE1* gene is the pea shape gene Mendel was tracking (Bhattacharyya *et al*., 1990). Even as of 2022, scientists have not identified the genetic locus for all seven traits that Mendel studied in the 1800s (Sussmilch *et al.*, 2022).
- In order to make this lab more accessible to introductory classes we omitted discussion of SBE1 protein function and the specific nature of the genetic differences between the *SBE1* alleles. These may be interesting topics of discussion in more advanced classes.
    - SBE1 stands for Starch Branching Enzyme 1. In the absence of functional SBE1 protein, more simple sugars remain in the pea instead of being incorporated into starch. The higher concentration of simple sugars causes more water to enter the pea while it is fresh and it swells. When these pea seeds dry out, they shrivel, giving a wrinkled appearance.
    - The mutation that causes wrinkled peas is an 800 bp transposon insertion in an exon near the end of the coding region of the *SBE1* gene (Bhattacharyya et al., 1990).

References

Bhattacharyya, M.K., Smith, A.M., Ellis, T.H., Hedley, C., and Martin, C. (1990). The wrinkled-seed character of pea described by Mendel is caused by a transposon-like insertion in a gene encoding starch-branching enzyme. *Cell* 60, 115–122.10.1016/0092-8674(90)90721-p.

Sussmilch, F.C., Ross, J.J., and Reid, J.B. (2022). Mendel: From genes to genome. *Plant Physiol* 190, 2103–2114. 10.1093/plphys/kiac424.

Learning goals and skills developed

Student Learning Goals - students will:

- Correlate genotype and phenotype
- Predict genotype and phenotype using Punnett squares
- Solve real-world problems using genetic analysis

Scientific Inquiry Skills - students will:

- Follow detailed experimental protocols
- Interpret data presented in a chart or table
- Use data to evaluate a hypothesis
- Make a claim based in scientific evidence
- Use reasoning to justify a scientific claim

Molecular Biology Skills:

- Micropipetting
- Principles of PCR
- Preparation of agarose gels
- Agarose gel electrophoresis

Standards alignment

Next Generation Science Standards

Students who demonstrate understanding can:

HS-LS3-1.	Ask questions to clarify relationships about the role of DNA and chromosomes in coding the instructions for characteristic traits passed from parents to offspring.
HS-LS3-3.	Apply concepts of statistics and probability to explain the variation and distribution of expressed traits in a population.

Science and Engineering Practice	Disciplinary Core Ideas	Crosscutting Concepts
• Asking Questions and Defining Problems • Analyzing and Interpreting Data • Constructing Explanations and Designing Solutions • Engaging in Argument from Evidence • Obtaining, Evaluating, and Communicating Information	**LS3.A: Inheritance of Traits** **LS3.B: Variation of Traits**	• Patterns • Cause and Effect • Interdependence of Science • Engineering, and Technology • Influence of Engineering, Technology, and Science on Society and the Natural World

Common Core ELA/Literacy Standards

RST.9-10.1	Cite specific textual evidence to support analysis of science and technical texts, attending to the precise details of explanations or descriptions.
RST.9-10.3	Follow precisely a complex multistep procedure when carrying out experiments, taking measurements, or performing technical tasks, attending to special cases or exceptions defined in the text.
RST.9-10.4	Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 9-10 texts and topics.
RST.9-10.5	Analyze the structure of the relationships among concepts in a text, including relationships among key terms (e.g., force, friction, reaction force, energy).
RST.9-10.9	Compare and contrast findings presented in a text to those from other sources (including their own experiments), noting when the findings support or contradict previous explanations or accounts.
WHST.9-10.1	Write arguments focused on discipline-specific content.
WHST.9-10.2	Write informative/explanatory texts, including the narration of historical events, scientific procedures/experiments, or technical processes.
WHST.9-10.9	Draw evidence from informational texts to support analysis, reflection, and research.

*For simplicity, this activity has been aligned to high school NGSS and grades 9-10 Common Core standards.