

Synthetic **biology**

TEACHER CURRICULUM

This curriculum has been developed by the iGEM team at The College of William & Mary. Activities have all been tried, tested, and enjoyed by students grades 1-12. If you have any questions regarding this curriculum, please do not hesitate to contact the iGEM team at igem@wm.edu.

WHAT IS

SYNTHETIC BIOLOGY?

From *SyntheticBiology.org*,
"Synthetic biology refers to both:
the design and fabrication of
biological components and
systems that do not already exist
in the natural world [and]
the re-design and fabrication of
existing biological systems."

Here at W&M iGEM, we don't think anyone should need to Google that definition. Synthetic biology is an incredibly interdisciplinary and powerful emerging field of science, and access to all that SynBio has to offer should not be left to students who happen stumble upon it in college. As synthetic biology becomes increasingly prevalent in our everyday lives, people should have the knowledge to recognize how SynBio will affect them. Unfortunately, surveys, including our own, show most people don't understand what synthetic biology is, but they are afraid of the consequences.

Our Solution

WORKSHOPS & ACTIVITIES

This summer, our iGEM team has held eight workshops across the state, both in our lab and at other schools, for students and parents alike. The feedback has been overwhelming. Following each event parents email us about how their children are still talking about the activities, and adults stay after to share their excitement about being able to openly learn and communicate with the people actually doing the science!

Sustainability

CURRICULUM & SUPPORT

Being on a summer iGEM team is an exciting part of any synthetic biology researcher's life, but the sad truth is that we will not be able to hold large workshops for students and parents throughout the school year. However, the learning should not stop with us. We have developed this curriculum so that anyone, with any degree of science background, can teach synthetic biology in an engaging way!

TABLE OF CONTENTS

A Twizzler DNA

GRADES 1-3

B DNA Bracelet

GRADES 1-3

C Tree of Traits

GRADES 1-3

F Monster Alleles

GRADES 2-5

E DNA Extraction

GRADES 3-10

F Codon Cards

GRADES 4-7

G Dessert Cells

GRADES 4-7

H Bean Genes

GRADES 5-8

I Egg Science

GRADES 5-8

J Yeast Streak

GRADES 5-9

K Soap Membrane

GRADES 5-9

L Electrophoresis

GRADES 5-10

M Cell Phases

GRADES 7-9

N Human Alleles

GRADE 9/HONORS

O Size of Things

GRADE 9/HONORS

P Plasmid Bracelet

GRADE 9/HONORS

Q Primer Design

HONORS

R Transformation

HONORS

S BacteriMeasure

HONORS

T if A, then T

HONORS/AP BIO

U RFP Art

HONORS/AP BIO

V Plasmid Design

AP BIO

W Manual PCR

AP BIO

X SynBio Ethics

AP BIO

Twizzler DNA

grades 1-3

WHAT DOES DNA LOOK LIKE?

time: 30 minutes



Learning Objectives

In this module, students will learn the basic structure of DNA. They will learn that DNA looks like a twisted ladder, and that it holds instructions 'written' in base pairs. They will understand that specific bases bind with other specific bases to form base pairs.

Materials

- Each participant will need:
- 2 Twizzlers
- 5 toothpicks
- 10 colored marshmallows*

*You will need 4 different colors. Each color represents a nucleotide base. You can use gummy bears instead of marshmallows.

Procedure

Introduce the concept of DNA (tips on how to do this are in the **Background**). Explain that the twizzlers represent the backbones of DNA, and ask them what they think DNA looks like (it is helpful to let them draw their guesses). Discuss how information may be carried by DNA through base pairs. Pass out marshmallows, assigning each color a base pair (adenine, thymine, guanine, and cytosine, or A, T, G, and C). Explain that A always matches T and G always matches C. Give each student one colored marshmallow and ask what color marshmallow they need to make a base pair, checking that they understand the concept of matching bases forming base pairs. Students should put two matching marshmallows onto each of the five toothpicks and attach them between the Twizzlers. Twist the whole structure to see the double helix.

Approximate Cost: \$6 for 25 students

Background

How does your body know to be human and not a dog or a jellyfish or a cactus? DNA! DNA, also known as deoxyribose nucleic acid, is a tiny molecule that every part of your body 'reads' in order to make you who you are. DNA tells your nose to be a nose and your foot to be a foot; it tells your hair what color to be and tells your skin if you have freckles. So... what is DNA? We know what it does, but what does it look like? If you had super vision that let you see the tiniest molecules and atoms, what would you see?

In this activity, we use Twizzlers to represent the backbone of DNA, because in the body the backbone is made of sugar - similar to the sugar in this candy - and phosphate, a yellow-brown mineral.

If DNA is a set of instructions, bases are like letters. The DNA alphabet only has four letters, A, T, C, and G. Two bases make a base pair. Explain that one marshmallow (representing one base) is attached to one backbone and the other marshmallow is attached to other backbone. If the colors of the marshmallows match, then they 'click' together and bind to one another, kind of like puzzle pieces.

Critical Thinking Questions

What happens if a DNA strand has A matched to C or G matched to T?

How many different DNA sequences did you see in your classroom?

If there were a billion base pairs instead of just five like you had on your model, how many different DNA sequences do you think you would see?

Do different people have different DNA sequences?

What kinds of information might a DNA sequence tell the body?

DNA Bracelet

grades 1-3

WHAT DOES MY DNA SAY ABOUT ME?

time: 30 minutes



Learning Objectives

This activity should engage students in examining the types of traits their DNA codes for. The bracelet concept also implies the sequential structure of genes: each bead represents a locus for the gene (order/location matters!). Students should notice that their bracelets look different from each other, because their DNA is different. If there are siblings in the class, their bracelets may look very similar, because genes are inherited.

Materials

- Colored pony beads: one bead per student per trait
- String/Pipe cleaners: one per student
- Key rings to make key chains instead of bracelets
- Traits table (see template)

Procedure

Each student will need a copy of the included **Traits Table**. The table lists questions about traits the student may have. All the questions have 'yes' or 'no' answers. Students should highlight or circle the 'yes' or 'no' column for each row. Within the Yes/No columns are colors. If the student answers 'Yes' for a trait, that student should add a bead of the corresponding color to his or her bracelet/keychain, and likewise if the student answers 'No.'

The beads should all be added in the same order as the traits table, to represent how each gene has its own specific location on a DNA strand.

Approximate Cost: \$8 for 25 students

Background

DNA is deoxyribonucleic acid. DNA molecules are very stable, very long, and composed of nucleotides.

Nucleotides include cytosine, adenine, guanine, and thymine, abbreviated respectively as C, A, G, and T. These nucleotides are “strung together,” one after another in a DNA molecule, forming a sequence (i.e. TACTAG... and on).

The sequence of nucleotides composing a DNA molecule forms a genetic code, which the cell can “read.”

DNA is structured as a double helix, two connected helices running in opposite directions. The two helices connect as the nucleotides align against each other. Cytosine nucleotides on one helix connect to guanine nucleotides on the opposite, and adenine connects to thymine. (C to G, A to T).

DNA codes for all biological aspects of the organism. Genetically determined traits include hair color, eye color, height, blood type, and sex.

Critical Thinking Questions

Knowing what you do about the two DNA backbones running in opposite directions and the way the nucleotides pair, can you predict the corresponding sequence on the opposite DNA backbone to “TACTAG” ?

What other traits do you think your DNA determines?

Do you think DNA determines all of your traits?

Tree of Traits

grades 2-4

ARE ALL TRAITS EQUALLY COMMON?

time: 45 minutes



Learning Objectives:

In this activity, students should explore the concept of genes. The traits on the included list all have yes/no answers (the student will have the trait or will not) and are meant to demonstrate examples of easily visible traits that our genes code for, and the diversity therein. Once class data is compiled, students should practice graphing the information. Finally, students should take some time to discuss in groups the implications of genes, and how they make everyone different (see **Critical Thinking Questions**).

Materials

- Graph paper
- Markers
- Ruler

Procedure

Students should highlight a box in the "Yes" or "No" column for each trait listed in the included traits table. In case you are doing this activity in conjunction with the DNA bracelet, each trait lists a color in the Yes/No column. These tables should be collected and the data graphed in histograms, showing the frequency of each trait.

By the end, students should be able to compare their tables, bracelets, and/or key chains. Who are they most similar to? Who are they most different from? Which traits are the most common? Which traits are most rare? Can you match a bracelet back to a person using the table? This is like using DNA to tell people apart!

Approximate Cost: free!

Background

The DNA inside every human contains instructions which help determine who they are--how their body starts out and how it will grow. DNA can be compared to an instruction manual. A gene is a specific DNA instruction for one small part of the body--for example, one gene determines whether someone has dimples or not, but it takes many genes to determine the color of someone's eyes. A gene can be compared to a sentence of the DNA instruction manual. It is important to know that genes do not always have the final say on how someone turns out--even if someone's genes have instructions for being of average height, environmental factors, like the quality of their diet and the frequency of exercise, can make them end up taller or shorter than expected. It is important to remember that genes are a very fundamental, baseline-level concept. In order for any life to function it needs some sort of information storage system to dictate how it functions and reproduces itself--genes are this information storage system. Please take caution not to instill the notion that genes are only responsible for minute, easily observable physical traits like those presented in this activity.

Critical Thinking Questions

What do you think would happen if there were no genes?
If you could change the instructions in your genes, would you?
What would you change them to say?
How do you think this would change the person you are today?
You can't control what the instructions in your genes say, and you can't change them. How does this make you feel about treating people differently based on the traits from their genes?

HELLO
MY NAME IS

and these are my traits...

Bead Number:	Trait	Yes	No
1	Are you female?	pink	blue
2	Do you have brown eyes?	red	white
3	Do you have blonde hair?	yellow	black
4	Do you have attached earlobes?	green	blue
5	Do you have a Window's Peak hair pattern?	white	yellow
6	Can you roll your tongue?	blue	black
7	Are you left handed?	orange	red
8	Do you have a round jaw?	yellow	green
9	Do you have a cleft in your chin?	red	black
10	Is your hair curly?	yellow	blue
11	Do you have big eyebrows?	green	yellow
12	Are your eyebrows straight?	white	black
13	Do you have short eyelashes?	blue	red
14	Do you have dimples?	orange	green
15	Do you have long fingers?	yellow	orange
16	Do you have freckles?	black	pink
17	Can you make a clover shape with your tongue?	red	yellow
18	Can you fold your tongue sideways?	green	white
19	Do you have a ridge on your nose?	red	pink
20	Do you have Hitch-Hiker's thumb?	black	orange
21	Do you sunburn easily?	white	yellow
22	When you clasp your hands together, is your left thumb over your right?	blue	white
23	Do you have big feet?	yellow	black
24	Do you have wide nostrils?	orange	pink
25	Are you colorblind?	red	green
26	Are you tall?	blue	orange

Monster Alleles

grades 2-5

WHAT ARE DOMINANT AND RECESSIVE GENES?

time: 30 minutes



Learning Objectives

Students should discover how some traits are more common than others due to dominant and recessive genes. They will learn about the concept of 'alleles'. Students will have the opportunity to practice converting genotype to phenotype through their monster drawings. They will see some generalized examples of the kinds of traits genes can code for.

Materials

- Colored crayons
- Paper
- Allele cards (see template)
- Allele key (see template)

Procedure

Before beginning:

Prepare allele cards (included in the template), and the allele key (also included in the template), or have students make their own allele card sets on index cards.

After cards are prepared:

Discuss the concept of alleles with students. Explain the meanings of dominant, recessive, homozygous, and heterozygous. Discuss the allele key with students: explain which traits the cards will decide for the monster they draw (i.e. a long tail is dominant over a short tail). Finally, shuffle the cards and lay them out on a table. Have students match all the letters and determine if the monster will be homozygous dominant, homozygous recessive, or heterozygous for each trait. Have the students draw their monsters based on the given traits. Point out that everyone's monsters still look very different-- that's because we only specified a few traits! A real monster's DNA would specify almost every trait. If we had cards for every trait, everyone's monsters would look very similar.

Approximate Cost: free!

Background

Alleles are different forms of the same gene. We are born with two sets of chromosomes. One set comes from our mothers, and the other set comes from our fathers. However, our mothers and fathers do not have exactly the same genes. For example, if a mother has blue eyes and a father has brown eyes, a baby will have two copies of the eye color gene that say different things. How does the baby's body decide what color to make the eyes?

The answer is in dominant and recessive genes. A dominant gene is represented by a capital letter, and recessive by a lowercase letter. We know that blue eyes are a recessive trait (b) and brown eyes are a dominant trait (B). If a baby has one copy that says blue and one copy that says brown (Bb, heterozygous), the baby will have brown eyes, because brown is dominant. If the baby had two copies of genes that said 'blue eyes,' the baby's eyes would be blue (bb, homozygous recessive), and if the baby had two copies that said 'brown eyes,' the baby's eyes would be brown (BB, homozygous dominant).

Critical Thinking Questions

How do individuals get their traits?

Do plants and animals have dominant and recessive traits too?

Bacteria reproduce by splitting off from themselves. Does this affect how they get their traits? (There are not two sets of chromosomes.)

How does this influence diversity in traits?

How could you change someone's traits?

Do two individuals with the same parents get all the same traits?

Monster Alleles Key

This activity uses cards (following pages) that should be printed such that the two pink pages are back to back, and the two green pages are back to back. The T should line up with the t on the other side of the page, so that when the cards are cut out, each card has a lowercase letter corresponding to the capital letter on the other side.

Alternatively, you can make own Allele Cards using index cards.

Below is a sample key of what the cards can be used to represent. Feel free to adjust the traits we have given, or make your own key, with whatever traits you would like the game to specify.

Dominant	Trait	Recessive	Trait
T	long tail	t	short tail
A	antennae	a	no antennae
D	droopy ears	d	pointy ears
G	beak	g	no beak
E	two eyes	e	one eye
H	horns	h	no horns
N	long neck	n	short neck
B	breathes fire	b	doesn't breathe fire
R	furry	r	scaly

Dominant	Trait	Recessive	Trait
T		t	
A		a	
D		d	
G		g	
E		e	
H		h	
N		n	
B		b	
R		r	

The cards will be mixed up, each color set represents one parent. Then, one card of each letter will be chosen from each parent to represent the alleles. If the combination is TT, for example, the monster will have a long tail. If the combination is Tt, the monster will still have a long tail, because the long tail is dominant. tt will call for a short tail, which is recessive.

A blank table is also provided if teachers would like to make their own key.

T

A

D

G

E

H

N

B

R

t

a

d

g

e

h

n

b

r

T

A

D

G

E

H

N

B

R

t

a

d

g

e

h

n

b

r

DNA Extraction

grades 3-10

HOW CAN WE TAKE DNA OUT OF A FRUIT?

time: 90 minutes



Learning Objectives

Students extract DNA from fruit to get a tangible sense of what DNA looks like on a macroscopic scale. They should understand that there is DNA in every cell and that DNA contains the instructions for life. They are also introduced to protocols that synthetic biologists use in order to research genetics and modify DNA.

Materials

Per student:

- ¼ of a banana*
- Ziploc bag with 1 inch of water
- Detergent
- Disposable cup with two coffee filters taped to the top
- Ethanol (refrigerate before use)
- One wooden skewer

*Any fruit can be substituted

Procedure

First, introduce the idea of DNA. *DNA is a tiny molecule that acts like an instruction manual for each of your cells. Your DNA tells you to be you, and a banana's DNA tells it to be a banana.* Let students guess what DNA looks like on a human scale (without a microscope).

Give each student (or pair of students) 1/4 of a banana in an open ziploc bag with water, and close the bag. Tell students to **gently** mash up the banana by squeezing the bag. Once the banana is mush and doesn't have any lumps, put some detergent into each bag, and close the bag. Students should gently mix in the detergent, trying to avoid creating bubbles

Give each student or pair a disposable cup with two coffee filters taped to the top. Carefully pour the contents of the bag into the coffee filters and let the liquid drip through the filter into the cup. Let the banana-detergent mix sit in the cup for 20-30 minutes. This is a good time to talk about DNA, or do a different activity from this collection, like codon cards or monster alleles.

Throw away the coffee filters and any material that didn't drip through the filter, keeping the cup and the liquid that collected in it.

Pour cold ethanol into cup. The more you pour in, the more DNA will precipitate out of the liquid. Use the toothpick or skewer to stir the white-clear substance that precipitates out, or collects at the top, of the liquid: this is banana DNA!

Approximate Cost: \$10 for 25 students

Background

DNA is located in the nucleus of the cell.

DNA contains the code for all biological aspects of an organism.

Individual genes are parts of the DNA that code to control particular traits.

Scientists extract DNA to study particular genes and how they affect organisms.

Scientists can study DNA in individual humans to determine traits relevant to their health and development and diseases to which they may be susceptible.

Scientists can change the expression of genes in organisms. This is most easily done in simple organisms like bacteria.

The process of changing genes involves a process called molecular cloning.

Molecular cloning is changing the composition of a DNA molecule and expressing that DNA in an organisms.

What each step of the extraction does:

Mashing up the banana allows the most possible banana cells to be exposed to the water and whatever liquid is added to the bag. The detergent breaks open the cells so that the DNA can enter the liquid solution. The filter removes the broken cells. The ethanol separates the DNA from everything else in the solution.

Critical Thinking Questions

Now that you know what banana DNA looks like, what do you think human DNA looks like? Why?

If you put banana DNA in a human cell, what would the cell look like?

If you could cut and paste pieces of banana DNA into apple DNA, what traits would you give the apple?

Would you eat fruit that had modified DNA? Why or why not?

How do you feel about the idea of putting DNA from one organism into another organism? What are the potential benefits of 'recombining' DNA? potential negative effects?

Codon Cards

grades 4-7

HOW DOES DNA MAKE AMINO ACIDS?

time: 30 minutes



Learning Objectives

Students should learn that there are four bases: adenine, thymine, guanine, and cytosine. A and T bind, and G and C bind. They should understand that a sequence of DNA has another specific sequence that it can bind to in order to create double-stranded DNA. Students will interpret their three base sequence, called a codon, and learn which amino acid their codon encodes.

Materials

- Class set of amino acid - codon chart (see template)
- Codon Card Key (see template)
- Codon cards - one match per two students (see template)

Procedure

First, introduce the idea of DNA and explain bases and base pairs. Then, tell students which color circle represents which base:

blue = adenine; red = thymine; green = guanine; yellow = cytosine

Pass out codon cards, face down so that students can't see the colored circles. Have students hold the cards up on their foreheads so that they can't see the card, but everyone else can - make sure that the orange triangle is pointing up. Tell students to ask other students yes or no questions about their sequence, ex. is my first base adenine / is my first circle blue? Once students think they know what is on their card, they should find a partner with the corresponding sequence. If a pair thinks they match, they should ask the teacher to confirm if they match or not - you can check the match by referring to key. After all the pairs are matched, explain that each three base pair sequence, called a codon, codes for one amino acid. Show students the amino acid - codon chart and ask pairs to find out what amino acid their sequence encodes (based on the coding strand: the one with the black line under the sequence). Then check if pairs identified their correct amino acid by using the key. Summarize what students learned: base pairs match, every three base pairs code for one amino acid.

Approximate Cost: \$2 for 25 students

Background

DNA is composed of nucleotides, including adenine, guanine, thymine, and cytosine, abbreviated A, G, T, and C. These nucleotides are “stringed together” forming a sequence in the DNA molecule. The sequence of nucleotides in the DNA compose a genetic code which the cell can “read.”

The cell first transcribes the DNA code by making another molecule, RNA, based on the code.

The cell then translates the RNA, using the sequence of the RNA, which comes from the DNA, to determine how to build proteins. Proteins compose an organism. Proteins are made of amino acids, which are “stringed together” forming a sequence in a somewhat similar way (conceptually, not chemically) to the way nucleotides are stringed together in DNA.

RNA is read in groups of three nucleotides. These groups are called codons. Each codon signals to add a particular amino acid to the protein for which a sequence codes.

Critical Thinking Questions

If you know a sequence of DNA, can you predict the corresponding sequence?

Why is there a coding strand and a non-coding strand? What would happen if both strands were read by the cell? Would they create the same amino acids?

Proteins are made of many amino acids. How could you change DNA to create a protein?

If one base in a sequence is deleted, what happens to the sequence following it? Would those sequences code for the same amino acids or would the codons be affected?

	T	C	A	G
T	TTT Phe TTC TTA Leu TTG	TCT Ser TCC TCA TCG	TAT Tyr TAC TAA Stop TAG	TGT Cys TGC TGA Stop TGG Trp
C	CTT Leu CTC CTA CTG	CCT Pro CCC CCA CCG	CAT His CAC CAA Glu CAG	CGT Arg CGC CGA CGC
A	ATT Ile ATC ATA Met ATG	ACT Thr ACC ACA ACG	AAT Asn AAC AAA Lys AAG	AGT Arg AGC AGA Ser AGG
G	GTT Val GTC CTA GTG	GCT Ala GCC GCA GCG	GAT Asp GAC GAA Glu GAG	GGT Gly GGC GGA GGG

Codon Cards

Key



Adenine



Guanine



Thymine



Cytosine



Coding Strand



Non-Coding Strand

Pair 1 - Threonine



A C G

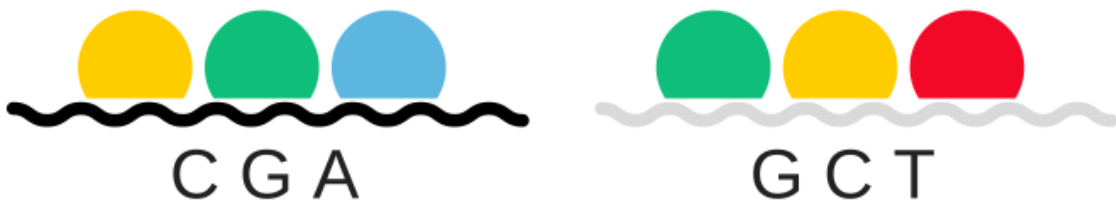


T G C

Pair 2 - Tryptophan



Pair 3 - Arginine



Pair 4 - Leucine



Pair 5 - stop codon



Pair 6 - Serine



Pair 7 - Tyrosine



Pair 8 - Arginine



Pair 9 - Glutamine



Pair 10 - Isoleucine



Pair 11 - Arginine



Pair 12 - Glycine



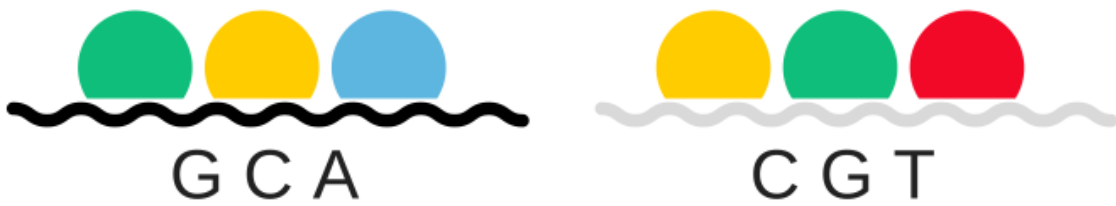
Pair 13 - Glutamic Acid



Pair 14 - Alanine



Pair 15 - Alanine



Dessert Cells

grades 5-6

HOW DO PLANT, ANIMAL, AND BACTERIAL CELLS DIFFER?

time: 60 minutes



Learning Objectives

Students should learn the organelles in a cell as well as the approximate relative size of each organelle. They should understand that there is no "empty space" in a cell. They should learn the main function of some organelles and understand that each organelle has a unique job within the cell and that the organelles work together to perform larger functions.

Materials

- Cookie
- Poptart
- Hotdog bun
- Spread (jam, butter, apple sauce)
- Assorted toppings to represent organelles

Procedure

With students, make a 3-way Venn Diagram of the similarities and differences in animal, plant, and bacterial cell structures. Discuss the role of each organelle.

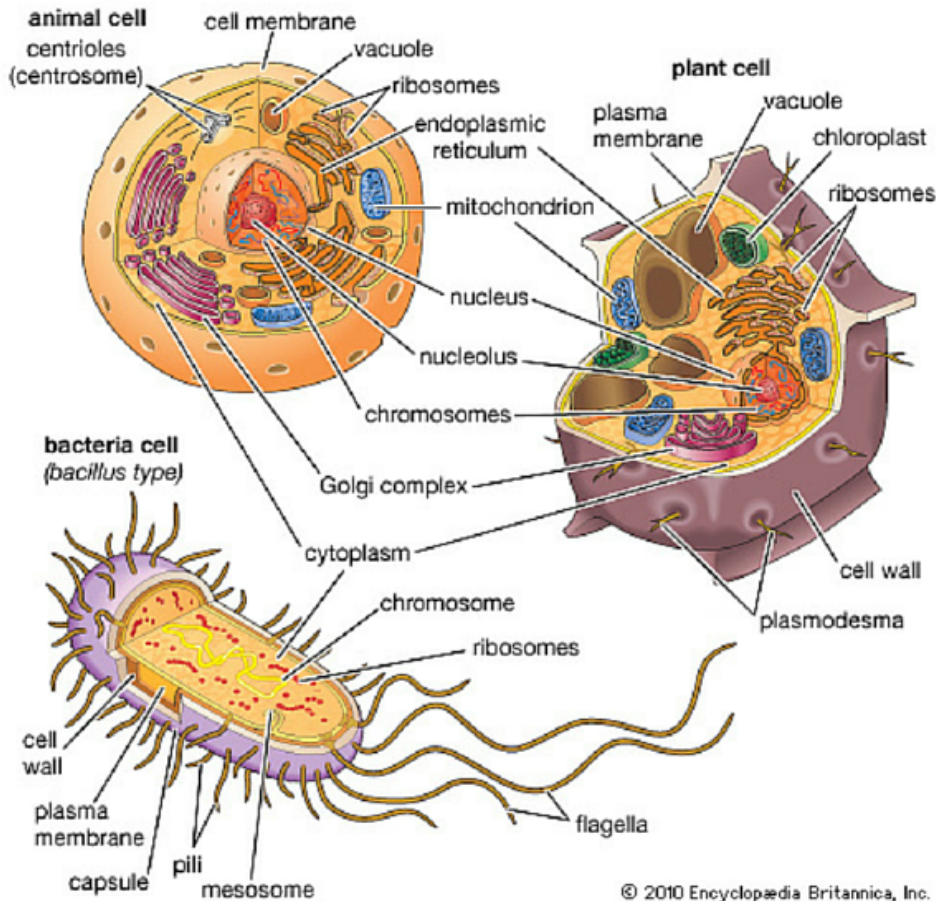
Then, pass one sugar cookie to each child to represent an animal cell, a piece of bread to represent a plant cell, and half a hotdog bun to represent a bacterial cell. Apply toppings to represent organelles and important cell structures that make each cell type unique. The same organelles should be represented by the same topping across different "cells". Try to make the 'organelles' roughly accurate size: the nucleus should be much larger than other organelles, the mitochondria and ribosomes should be small and there should be many of them, etc.

After students complete their 'cells' discuss which toppings represent which organelles and why.

Approximate Cost: \$15 for 25 students

Background

Some typical cells



Plant and animal cells are eukaryotes because they have nuclei. Bacterial cells are prokaryotes because they do not have nuclei. Plant cells and bacterial cells both have cell walls, but animal cells do not.

Bacterial cells have pili and flagella to help them move and sense, animal and plant cells do not.

Critical Thinking Questions

Why do bacteria need flagella, but animal and plant cells do not?

Why would it be like if animal cells and cell walls?

Which cell would it be easiest to add/change DNA in?

Which cell type seems most primitive? Which seems most advanced?

Dessert Cells

For each organelle write the function and what you used to represent the organelle.

Nucleus

Represented by: _____

Function: _____

Mitochondria

Represented by: _____

Function: _____

Cytoplasm

Represented by: _____

Function: _____

Ribosomes

Represented by: _____

Function: _____

Rough ER

Represented by: _____

Function: _____

Smooth ER

Represented by: _____

Function: _____

For each organelle write the function and what you used to represent the organelle.

Golgi Body

Represented by: _____

Function: _____

Cytoskeleton

Represented by: _____

Function: _____

Vacuoles

Represented by: _____

Function: _____

Peroxisome

Represented by: _____

Function: _____

Lysosomes

Represented by: _____

Function: _____

Bean Genes

grades 5-8

WHAT ARE INCOMPLETE DOMINANCE AND CODOMINANCE?

time: 45 minutes



Learning Objectives

Students will learn about heredity and how genes can combine. It is an excellent introduction to Punnett Squares as students will explore the ratios among phenotypic expression. Students will develop a deeper understanding of homozygous, heterozygous, dominant, recessive, and learn about codominance and incomplete dominance.

Materials

- Per group of students:
- (recommended pairs)
- 10 red beans
- 10 white beans
- 2 paper bags
- Garden Tally sheet (included)

Procedure

Give each pair/group of students 2 paper bags. In one bag put 10 red beans, and in the other, put 10 white beans. The beans represent the genes for flower color (red or white). Red flowers are dominant (R), and white flowers are recessive (r). Choose one bag to be the female flower and the other to be the male flower: then, pick one bean from each bag with your eyes closed. If both are red (RR) color a flower on the **Garden Tally** red. If both are white, leave a flower white (rr). If one is red and the other is white, color the flower pink (Rr) for incomplete dominance or spotted red/white for codominance (one partner should do codominance, the other should do incomplete dominance). Return each bean to its original bag. Keep going until all your flowers have been accounted for. Be very careful that each bag always has 10 of the correct color bean!

When students are done, they should tally their results in the red boxes. Which color flower was most common? What percentage of the total flowers were that color? What about least common?

Approximate Cost: \$5 for 25 students

Background

This module assumes basic understanding of recessive, dominant, homozygous, and heterozygous traits. If you would like further background information on these topics, please see the Monster Alleles background information.

The new information in this module is about incomplete dominance, and how it differs from codominance.

Incomplete dominance is when one allele for a trait is not entirely dominant over the other allele. The trait is still controlled by a single gene (with two alleles, one from each parent), and each of those alleles contributes equally to phenotypic expression.

In the context of flowers: pink flowers (the offspring of a red flower and a white flower) with the genotype Rr are an example of incomplete dominance. The phenotype is in between red and white.

What if the red and white genes were instead codominant? We will again see a third phenotype, but in this case, both parental traits appear together. The Rr flowers would be spotted red and white!

Another example of codominance is blood type: a mother with Type A and a father with Type B will produce offspring with Type AB: both A and B markers are on the surface of the offspring's red blood cells.

Critical Thinking Questions

If you cross two pink flowers, what are the possible phenotypes of the offspring?

Codominance and incomplete dominance both produce a third phenotype. Can two alleles produce more than 3 phenotypes?

HELLO

MY NAME IS

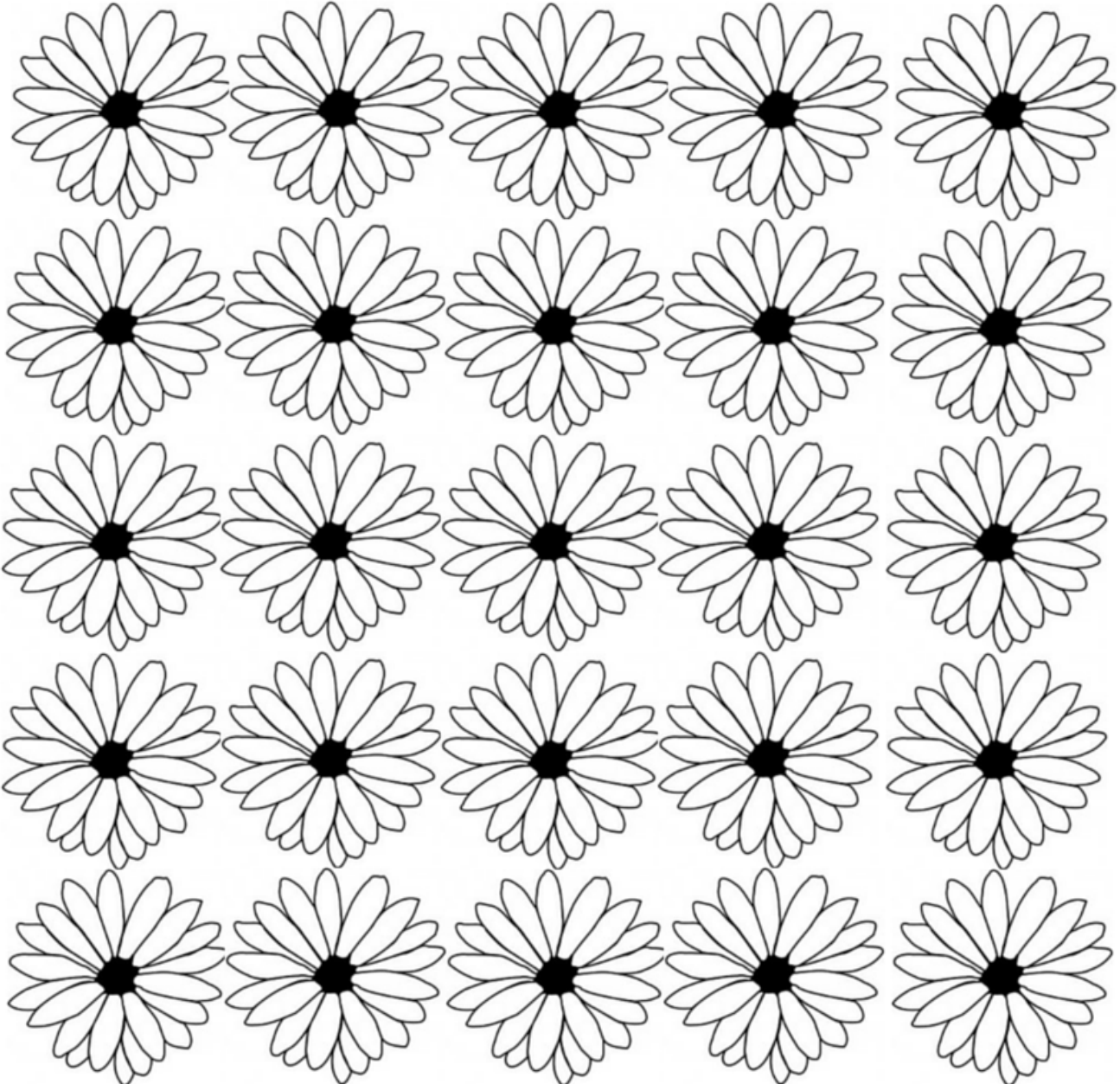
Blank space for writing a name.

Circle One:

Codominance

Incomplete Dominance

and this is my flower garden...



How many RR? %?

These should be red.

How many Rr? %?

Codominance: spotted red/white
Incomplete dominance: pink

How many rr? %?

These should be white.

Egg Science

grades 5-8

WHAT DOES THE SHAPE OF A PROTEIN DO?

time: 45 minutes



Learning Objectives

Students will explore how protein form follows function. This concept is vital in high school biology classes, and easy to visualize with the activity described here. Students will see how proteins can be heated, beat, and mixed with other solutions to change their structure and their outward appearance.

Materials

- A dozen eggs
- Hot plate
- Pot
- Bowls
- Frying pan
- Water
- Oil
- Whisk

Procedure

Unless you have a lot of hot plates, this is best done as a full class activity. Crack several eggs into three different bowls. Try heating the eggs in one of the bowls: put it on a frying pan and over a hot plate. How does the look of the egg change? Are there differences in consistency? Does the egg feel different? What about if you beat the egg?

In the second bowl, mix the egg with a whisk. What changes do you see? Finally, prepare an oil/water mixture. Discuss how the differences in oil/water polarity do not allow the two to mix. Then add egg to the oil/water solution, and watch as it creates an emulsion. Discuss how the egg proteins may be interacting with the oil and water to bring this about (you can alternatively use this activity to make pavlova, hollandaise sauce and deviled eggs).

Approximate Cost: \$4 for 25 students

Background

Proteins are made up of long chains of amino acids decided by your DNA. In the white part of an egg, we see 'globular' proteins: if you magnified them they would look like a long string folded up into a spherical shape. The proteins stay curled up tightly using many different types of chemical bonds.

When we heat the egg, the proteins become more energetic: they bounce into each other and into the water molecules (also in the egg). This breaks up some of the bonds that keep the proteins in their globular shape, and it also makes new bonds connecting the proteins together! If you keep heating, the egg proteins will form a web of interconnections, and your egg will feel rubbery.

When we beat the egg, like chefs do to make soufflé, air bubbles get caught in the egg mixture. Egg proteins have both hydrophobic (water-repelling) and a hydrophilic (water-attaching) ends. When the air gets caught in the egg, the hydrophobic part of the egg protein touches the air bubble, and the hydrophilic part stays in the water. However, the protein was all coiled up before. To make it so all the water-attracting parts can touch the water, the proteins uncurl, just like they did when we heated the egg. If you cook a whisked egg, the proteins will form connections with each other but leave the air bubbles in place: the egg will come out fluffier.

Finally, when you mix the egg into oil and water, the same hydrophobic/hydrophilic principle applies. The hydrophilic end of the protein attracts the water and the hydrophobic end attracts the oil, so water and oil molecules can be held close together with an egg protein in between.

Critical Thinking Questions

What foods can you think of that use the properties of egg proteins to affect texture?

Yeast Streak

grades 5-9

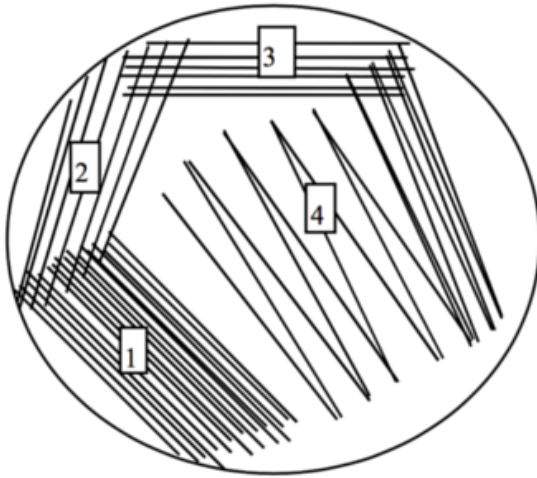
HOW CAN WE GROW FUNGAL COLONIES?

time: 45 minutes



Learning Objectives

Learn to make medium to grow colonies, pour plates, streak plates, and observe growth.



Materials

- Petri dishes or foil cupcake liners
- Gelatin medium (recipe in **Background**)
- Baker's yeast packet dissolved in 2 cups warm water
- Q-tips
- Each student should bring a vegetable or cheese (blue cheese works well)

Procedure

Heat the gelatin/agar solution and help students each pour 3mm of the solution into two petri dish. Allow these to solidify and label each plate with each student's name before proceeding.

Once the plates are solid, allow each student to dip the end of a toothpick into the yeast solution, and very gently glide the tip of the toothpick along one of the plates in the pattern shown above under **Learning**

Objectives. Try not to poke into the gelatin surface.

Next, allow students to do the same with a moist cotton swab run over a vegetable /cheese.

Use the cotton swab the same way as the tooth pick to streak the second plate. Allow plates to grow at room temperature for 4-7 days, and then observe.

Approximate Cost: \$15 for 25 students

Background

For the homemade medium, you will need to combine:

8 teaspoons sugar, 4 beef bouillon cubes, 4 envelopes of plain gelatin and 4 cups of water in a saucepan. Bring the solution to a boil as you stir. Allow it to cool slightly before allowing students to help pour plates. This recipe makes about 25 plates. Double the recipe so each student can have two plates. This mixture should provide the nutrients yeast and fungus need to grow.

Why do we streak plates in this pattern? Often, scientists would like to get distinct colonies on a plate. This is because each colony represents a spot where a single yeast cell, fungus cell, or, as synthetic biologists are often concerned with, bacterium, began replicating. All the cells in that colony are clones of the original cell. Area 1 on the streaks will be the most dense: colonies may not be distinct. We keep going with the toothpick/cotton swab so that eventually the colonies will be far enough apart, because there are fewer and fewer cells left on the toothpick/cotton swab as we zigzag more across the plate.

Critical Thinking Questions

If a scientist is trying to add a new piece of DNA to a bacterium, say, to make it fluoresce red, how can the scientist make sure only bacteria that have that gene grow on a plate? (Think about antibiotics.)

When a scientist grows bacteria on a plate and then extracts DNA from a single colony (so that all the DNA is the same, because they are all clones), is it better to do that extraction after the bacteria have grown for one day or after they have grown for one week? (Think about mutations)

Soap Membrane

grades 5-9

WHAT ARE THE PROPERTIES OF A CELL MEMBRANE?

time: 60 minutes



Learning Objectives

Learn about the properties of cell membranes through soap bubbles! The following properties will be covered:

1. Cell membranes are fluid and flexible. They can reform when broken.
2. Channel proteins float in the cell membrane. Channel proteins provide channels through the membrane.
3. Organelles are membrane-bound.
4. Binary fission.

Materials

- Corn Syrup
- Water
- Trays/Pans
- Bendable Straws
- Thread
- String/Twine

Procedure

Bend the straws, and crimp the short ends. Arrange them in a square to make the bubble frame. Tie thread in a small loop. Make bubble solution with 900mL water, 100mL dish soap, and 25mL corn syrup. To demonstrate each objective:

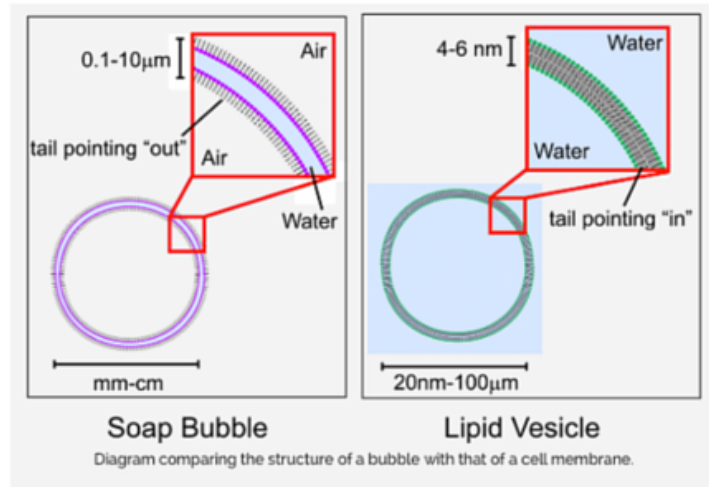
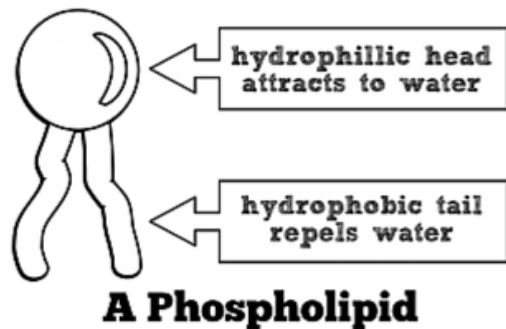
1. Dip bubble frame in solution. Move wet hand slowly through the soap film, watch as the film flexes and reforms when you take your hand out.
2. Put the thread loop in the bubble frame, and pop the part of the bubble inside the loop to represent a channel protein. Move the 'channel' around.
3. Use a straw to blow bubbles inside other bubbles.
4. Use the twine to split bubbles on the tray into multiple bubbles.

Approximate Cost: \$5 for 25 students

Background

The membrane properties demonstrated by the soap bubbles are described in the Learning Objectives.

Additionally, it is useful to know the similarities between soap and membranes. Cell membranes are made of phospholipids, which have polar heads that interact with water, and non polar tails that are hydrophobic. The phospholipids line up tail to tail, so that the inside of the membrane is shielded from water while the outside touches water. Soap bubbles are similar, but have a tail pointing outward and a layer of air in the middle.



Critical Thinking Questions

Why are membranes so important to life?

Can you think of anything else that acts like a membrane the way soap does?

Why do cell membranes have the hydrophobic tails facing in instead of out?

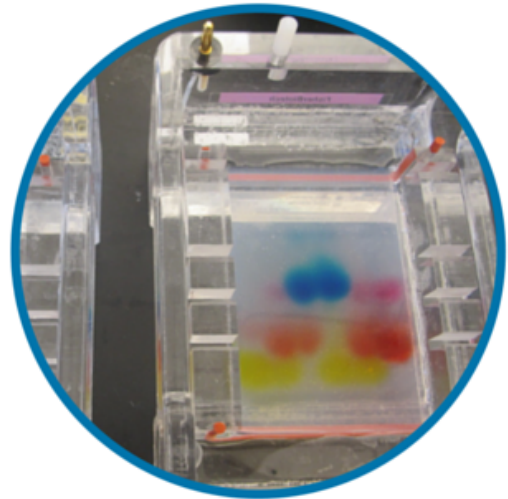
What other membrane properties can you think of that are not illustrated here? How could you illustrate them with bubbles?

Electrophoresis

grades 5-10

HOW CAN WE SEPARATE MOLECULES BY SIZE?

time: 60 minutes



Learning Objectives

Gel electrophoresis is used almost daily by many molecular biologists. Students will firsthand make a gel with wells to load, choose colors of food coloring to load the wells with, and load them in with a disposable pipette. With a teacher's help, they will run the colors down the gel using 5 9-Volt batteries, and watch as the colors separate based on molecular size. The same color will always split into the same bands. This separation corresponds to the way scientists separate DNA molecules by size.

Materials

- 5 9-Volt batteries
- 2 clamp wires
- Saltwater solution
- Gelatin/agarose
- Food coloring
- Disposable pipettes
- Plastic forks
- Tape

Procedure

Combine 50mL TAE buffer solution with 0.5 grams agarose, heat in the microwave, and pour into the provided gel frame with the fork-comb. Once solid, move the gel frame into the Tupperware container. Remove the fork-comb. Pour just enough TAE buffer to cover the gel and fill all the wells.

Finally, have students load the gel with 1 drop of food coloring in each well using disposable pipettes. For more colors, mix food coloring in advance.

Have students guess what colors their lane will split into, and which molecules they think may be heaviest/go the shortest distance. Once the gel is loaded, attach the 5 9-Volt batteries to positive leads, and lay these leads in the gel chamber solution, such that the positive lead is at the bottom (opposite to the wells, and the negative lead is at the top (close to the wells).

Approximate Cost: \$25 initially, reusable!

Background

A gel is best thought of as a dense web through which we are running small molecules. These molecules have a slight negative charge, thus the current will pull them toward the positive end of the batteries (toward the positive lead). However, food coloring colors are made up of the primaries (red, blue, and yellow-- some food coloring has a bit of pink that separates from the red as well). Each of these colors is a different size, so it moves at a different speed down the gel. Imagine a large molecule trying to move through dense web vs. a small molecule trying to move through the same web. Which will go further?

Scientists use electrophoresis to check the sizes of their DNA, because they cannot visually see how big the DNA molecules are. Usually, scientists know how long a piece they are looking for, and running a gel can help them understand if the piece they have is approximately the right size.

When scientists run gels using DNA, they also run a ladder in one of the wells, which has a bunch of DNA fragments of different, known sizes. That way, they can compare the band their DNA makes to the bands on the known ladder and estimate the size.

Critical Thinking Questions

What are some applications of gel electrophoresis?

How does the current separate the DNA fragments by size?

If we were using DNA instead of food coloring, it would be clear.

Scientists use a chemical called ethidium bromide in their gels, and then put the gel in a UV chamber to see where the DNA is. What does ethidium bromide do? Why might it be dangerous to us?

Why does DNA migrate toward the positive end? (Think about the charge on DNA molecules.)

Cell Phases

grades 7-9

HOW DO CELLS REPLICATE?

time: 60 minutes

Learning Objectives

Students will learn the phases of cell replication.



Materials

- 6 sugar cookies per participant
- Frosting/Peanut butter
- Food coloring
- Small candy of different shapes and sizes
- Sprinkles of different shapes and sizes

Procedure

Before beginning the activity, discuss with students the different phases of cell division (these phases are briefly outlined under **Background**).

Approximate Cost: \$8 for 25 students

Background

Multicellular organisms grow by cell division. Likewise, populations of unicellular organisms grow by cell division. When a cell divides, it must copy its DNA to pass to the new “daughter” cell. DNA is copied and the cell divides in a process called “mitosis.” (Mitosis refers to the division of cells in the production of somatic cells, which are all cells that are not gametes, neither sperm nor egg. In gamete production, cells divide by meiosis, which halves the number of chromosomes.)

Prophase: Chromosomes replicate

Prometaphase: Nuclear envelope dissolves, mitotic spindle (microtubules) attach to centromeres

Metaphase: Chromosomes line up along the middle.

Anaphase: Chromosomes divide and the microtubules shorten, pulling the chromosomes toward the centrosomes to either side of the cell. One copy of each chromosome goes to each side.

Telophase: A new nuclear envelope surrounds the sets of chromosomes at either end of the cell. Cytokinesis, the process by which the cell pinches together and forms two distinct cells, separates the two halves of the cell containing the two copies of the chromosomes. This forms the two daughter cells, which each hold an entire copy of the DNA.

Critical Thinking Questions

What would happen if, by mistake, a chromosome made a copy of itself, but failed to split when the centrosomes pulled during anaphase? How would this affect each of the daughter cells?

One cell would receive two copies of that chromosome, the other none. This type of error (of greater consequence when occurring meiotic (sex cell) division, because the error is then present in all cells of the child) is responsible for a set of genetic disorders that includes Down Syndrome caused by incorrect number of chromosomes.

Sex cells are produced by a process called meiosis, in which, essentially, there are two rounds of division for one copying of the chromosomes so that the number of chromosomes per daughter cell is halved. Rather than pairs of chromosomes holding two different versions of the genes, each daughter sex cell has one chromosome out of that pair. Why would it be necessary to halve the number of chromosomes for sex cells and not for other cells?

Sex cells unite during fertilisation, each bringing one set of chromosomes. This results in the normal paired set of chromosomes in the child.

Human Alleles

grade 9 & 11

HOW DOES A PUNNETT SQUARE WORK?

time: 60 minutes



Learning Objectives

This activity is a more detailed version of the monster alleles activity for younger students. Biology students should select traits for parents using the given cards and key, calculate Punnett Square probabilities for each trait in the offspring, cross the parental alleles by drawing them out of a hat, and then draw the offspring, listing the probability that the offspring would have each of the traits it was born with.

Materials

- Human Allele Cards (see template)
- Human Allele Key (see template)

Procedure

Choose traits of parents using the human allele cards for each parent. Have students draw Punnett Squares using the parents' genotypes to predict the child's phenotypes. Finally, using the allele templates, have each student cross the parents and draw the offspring. Each student should also write the probability of the child getting each of the traits he or she got based on the Punnett Square calculations.

Approximate Cost: \$8 for 25 students

Background

A Punnett Square is a simple representation of the potential genotypes of the offspring of two individuals with known genotype. Suppose we wanted to know the probability that the child of two people would have dimples. We use two letters to represent one allele, representing the two chromosomes: a capital letter represents a dominant trait and a lowercase letter represents a recessive trait. In this case, D would represent having dimples and d would represent a lack of dimples. Suppose both parents are heterozygous (Dd). Then you can construct a Punnett Square with one parent on each side.

	D	d
D	DD	Dd
d	Dd	dd

Now, just fill in the inside of the Square by taking the letter from the row and the letter from the column of that space. One is homozygous dominant (DD), two are heterozygous (Dd), and one is homozygous recessive (dd). This means 3 of the genotypes have a **dominant** phenotype and 1 has a **recessive** phenotype. Since there are 4 possible choices, this means there is a 3/4 chance that the child will have dimples and a 1/4 chance that it will not.

Critical Thinking Questions

When do you think the Punnett Square's prediction would be most accurate? When would it not be?

What about the Punnett Square's design makes it inaccurate in this situation?

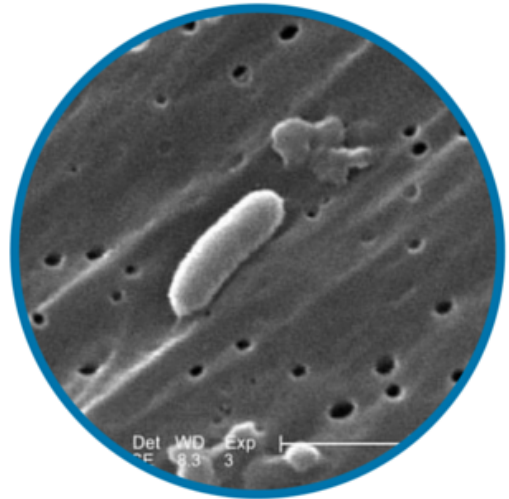
It seems like recessive phenotypes always have a lower probability of being selected for. Why don't recessive traits disappear from the population after a while?

The Size of Things

grade 9 & 11

WHAT SCALE ARE WE WORKING WITH?

time: 30 minutes



Learning Objectives

Students will develop a grasp of the relative sizes of microscopic levels of life. They should be able to better visualize the scale of cells and molecules like DNA. They should also understand that the size of things matters, especially to microbiologists. This activity also serves as an introduction to the quantitative size of biology.

Materials

- Computer with internet access
- Size of Things worksheet for each student (see template)
- Size of Things worksheet key

Procedure

Hand out the worksheet. Students should read the paragraph at the top of the worksheet. Before students answer any questions or fill in the chart, discuss with students why numbers are important in science. Then either on their own or as a class, students should find the numbers necessary to fill in the chart on the worksheet. All measurements should be in meters or other metric length units. Refer to the worksheet key if students have trouble finding the numbers online. For the one meter 'thing' students should choose anything they are familiar with that is about one meter, which is about a yard. If students have troubles with the calculations, walk them through the calculations on the key. After students complete the worksheet, ask them the critical thinking questions and discuss their answers with them.

Approximate Cost: free with a computer!

Background

Relevant background information is provided on the websites listed on the handout.

Critical Thinking Questions

Why do scientists use numbers?

Why does the size of DNA matter?

Why is it a good idea for DNA to be very small?

What would happen if DNA was bigger and cells were smaller?

Scientists use a technique called "Gel Electrophoresis" to learn the Length of DNA fragments. How would knowing the length of a DNA sequence be useful if scientists were trying to attach two pieces of DNA to each other?

Do you think banana DNA and human DNA are the same width? the same length? why?

The **Size** of Things



How many centimeters long is a pencil?

You probably don't know off the top of your head, but you probably do know that the length of a standard number two pencil, 7 1/2 inches, is similar to the length of your face.

Quantitative measurements are useful because they let us compare the relative size of things, even if those things are very far from each other in time or space. The sizes of things are especially important in science, where researchers strive to identify the causes of differences and similarities between things.

How big do you think a human skin cell is (write a number and unit)?

What is the smallest number of skin cells you think you can see with your bare eyes?

How wide do you think DNA is (from backbone to backbone)?

How long do you think DNA is?

If you stretched out all the DNA in a cell would it be taller or shorter than you are?

Using a computer, go to 'The Scale of the Universe 2' at

<http://htwins.net/scale2/> and 'BioNumbers' at

<http://bionumbers.hms.harvard.edu/KeyNumbers.aspx?redirect=false>

and fill in the following chart.

Size of Thing (m)	Thing
	Height of an Average Adult Human
	Length of a Human Skin Cell
	Width of DNA
	Length of DNA (per base pair)

How many base pairs of DNA would it take to span the length of a human skin cell?

How many human skin cells would it take to span a meter?

How many base pairs of DNA would it take to span a meter?

How many base pairs of DNA would it take to span the height of an average adult human?

There are about 3 billion base pairs of DNA in a human cell. Is all the DNA in a cell, stretched out, taller or shorter than you are? Does this match your prediction?



The **Size** of Things **Key**

Size of Thing (m)	Thing
1.7 m	Height of an Average Adult Human
3.5×10^{-5} m	Length of a Human Skin Cell
3×10^{-9} m	Width of DNA
3.3×10^{-10} m	Length of DNA (per base pair)

How many base pairs of DNA would it take to span the length of a human skin cell?

$$(3.5 / 3) \times 10^{(-5 - (-9))} = 1.17 \times 10^4 \text{ base pairs (bp)}$$

How many human skin cells would it take to span a meter?

$$(1/3.5) \times 10^{(0 - (-5))} = 0.296 \times 10^5 = 2.96 \times 10^4 \text{ cells}$$

How many base pairs of DNA would it take to span a meter?

$$(1/3) \times 10^{(0 - (-9))} = 0.333 \times 10^9 = 3.33 \times 10^8 \text{ bp}$$

How many base pairs of DNA would it take to span the height of an average adult human?

$$(3.33 \times 1.7) \times 10^{(8 + 0)} = 5.66 \times 10^8 \text{ bp}$$

There are about 3 billion base pairs of DNA in a human cell. Is all the DNA in a cell, stretched out, taller or shorter than you are? Does this match your prediction?

Taller: $3 \times 10^9 > 5.66 \times 10^8$



Plasmid Bracelets

grade 9 & 11

WHAT DO THE PARTS OF A PLASMID DO?

time: 45 minutes



Learning Objectives

In this module, students will discover the key components of a plasmid, and how they can be used to express genes from other animals. The bracelet/keychain made in this activity very closely models one of the parts, a two reporter system, that the W&M iGEM team is submitting to the registry this year. For further discussion on the two reporter system for extension of this activity, see wmigem.weebly.com. This module provides a strong introduction to synthetic biology.

Materials

- Key rings to do keychains instead
- Colored pony beads
- String/Pipecleaners

Procedure

Teachers should discuss the role of a plasmid before beginning this activity. There is the option to use beads for the backbone or to use a pipe cleaner and have the bare pipe cleaner be the backbone. Either way, there will also be beads representing each of the key regions: a promoter, ribosome binding site, gene for cyan fluorescent protein, gene for yellow fluorescent protein, and a double terminator. This structure is homologous to the GERALD part on the team blog, linked in **Learning**

Objectives. The sentence analogy (described in **Background**) is an excellent way to explain plasmids to students who have not worked with them before. Choose a color bead for each region (use extra of the same color for the genes as they are larger regions of DNA) and string them onto the bracelet/key chain in order. Tie it into a loop like a plasmid.

Approximate Cost: \$8 for 25 students

Background

A plasmid is a circular piece of DNA inside a bacterial cell. Bacteria do not have nuclear envelopes, so plasmids are transcribed/translated just like chromosomal DNA. Plasmids are very useful to bacteria for evolving. They can use horizontal gene transfer to share genes with one another. This helps bacteria adapt and mutate quickly under changing environmental conditions.

It is helpful to think of a plasmid as a sentence. Every sentence has an indication that it has begun (a capital letter), information it tells you (the words in the sentence, and a signal that the sentence has ended (a period). The capital letter is similar to the promoter, which signals to the bacterium to start reading DNA there (transcription). The ribosome binding site can also be compared to the capital letter at the beginning of a sentence, as this is where translation begins. The words in the sentence, or information passed on, is similar to the genes the plasmid codes for, for example cyan and yellow fluorescent proteins. The period is analogous to the double terminator, which signals the end of transcription and translation.

Critical Thinking Questions

Why can't plants and animals do horizontal gene transfer?

If a bacterium is expressing genes for both cyan and yellow fluorescent protein, what color will the bacterium appear under a confocal microscope? What about if the bacterium is expressing genes for both red and green fluorescent protein (remember, these are colors of light, not pigments, so they mix differently!)?

If all the bacteria on a plate are expressing each fluorescent protein to different degrees, how will the bacteria look as a whole? (see our blog)

Primer Design

H Biology

HOW CAN WE COMBINE AND AMPLIFY DNA?

time: 90 minutes



Learning Objectives

An overarching goal of this curriculum is to expose students to the same concepts and techniques that take place on a day-to-day basis in a biology lab. Primer design is not a protocol, persay, but is certainly a prerequisite to being able to develop any kind of biological device. This module should emphasize base-pairing rules, and introduce ideas about how different piece of DNA can be joined together (recombinant DNA), as well as how DNA regions can be amplified (PCR).

Materials

- Use the primer design worksheet (included)
- Computer with internet access

Procedure

Each student should use the worksheet provided and follow along on a personal computer to design primers for the given objectives. The worksheet provides step-by-step instructions, but we recommend that teachers try it once beforehand, in case any technical difficulties arise!

Approximate Cost: free with a computer!

Background

DNA polymerase can only add to the 3' end of DNA. However, scientists are often looking for ways to add pieces of DNA into the middle of a DNA strand, or to amplify a piece of DNA in the middle of a strand. In these situations, primers are extremely useful.

A primer is a short sequence of nucleotides that attaches to the template strand by base pairing. From here, DNA polymerase can bind to the 3' end of the primer and add nucleotides from there. Thus scientists can choose exactly where DNA polymerase starts.

It is important to remember that DNA strands are antiparallel: one side runs 5' to 3' and the other runs 3' to 5'. Because DNA polymerase only runs 5' to 3', we need two primers, one for each 5' side.

Critical Thinking Questions

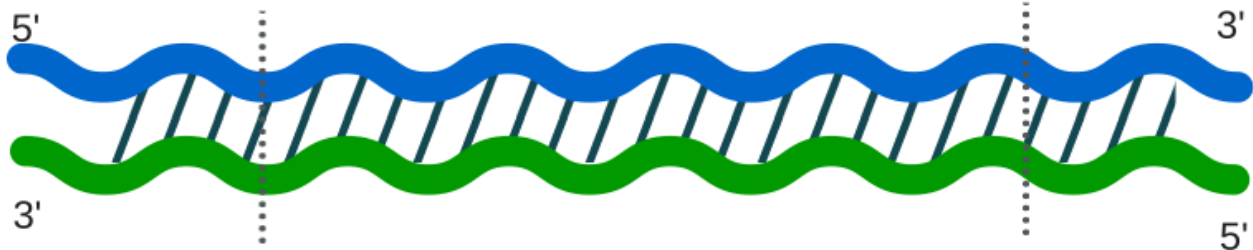
What would happen if primers are complimentary to each other?

Is it better to use longer primers or shorter primers?

When doing PCR, we use primer melting temperatures to determine what annealing temperature to use for amplification. What happens if the annealing temperature is too high (think about Taq polymerase, a sensitive enzyme).

Designing PCR Primers

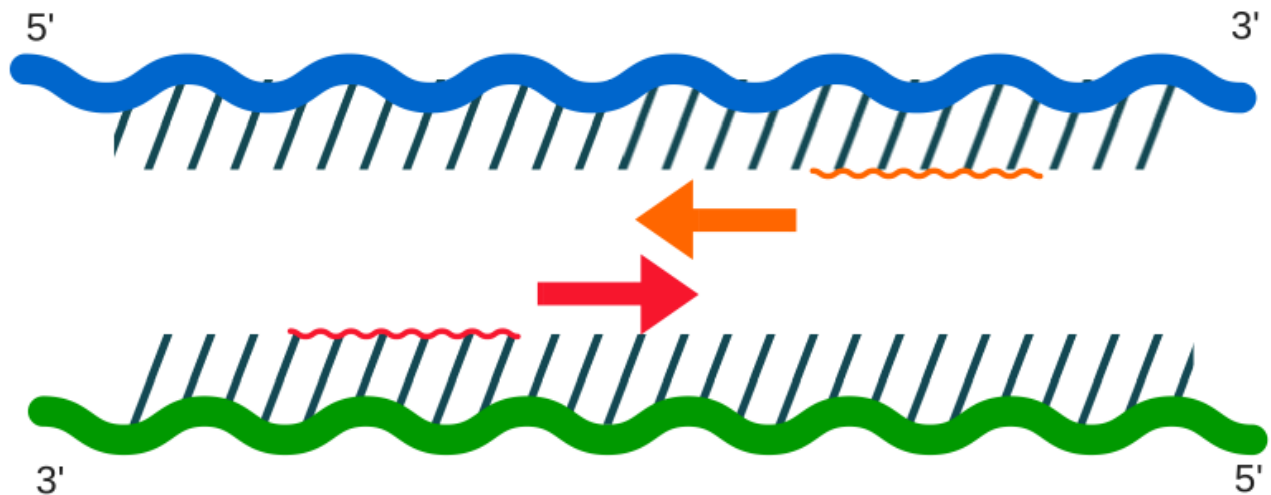
Designing primers for polymerase chain reaction (reference the PCR Process worksheet under 'Manual PCR' to learn how PCR works):



The region of DNA that we are trying to amplify is shown between the dotted grey lines.

Recall that DNA only amplifies from 5' to 3': that is, DNA polymerase can only add nucleotides to the 3' end of each single strand of DNA. Therefore, we will need two different primers!

The first primer, in orange, should bind to the far right of the target region. The second primer, in red, should bind to the far left of the target region.



From this point on, a polymerase will extend the region the primer bound to until the 5' end on each strand. Remember, DNA strands run antiparallel: the 5' end of the primer is on the same side as the 3' end of the template strand it is bound to. The orange and red arrows show the direction in which DNA polymerase will add nucleotides.

Now it is your turn. Before we can start designing primers, there are a few things we have to figure out. This worksheet will walk you through those questions.

Say we have the following sequence (representing the blue strand):

ATCGAATCCTAGTAAGCTCTAGTACTTGAGGTATCTCGACTTTAGCATGAAT

What is the sequence of the of the green strand?

The target region we want to amplify (from the blue strand) is as follows:

TCCTAGTAAGCTCTAGTACTTGAGGTATCTCGACTTTAG. **Highlight the complementary region in your green strand sequence above.**

We are almost ready to design our primers. Why is it that scientists don't just design a primer to span the whole sequence, rather than putting polymerases to work for extending the primers? The answer is that primer size really makes a difference! The length and sequence of a primer governs its T_m (melting temperature, which is used in calculating the annealing temperature for PCR), and the longer the primer, the more specific it is.

The melting temperature (T_m) of the primers, minus about 2 degrees C, is generally accepted as the annealing temperature for PCR. A rough formula for calculating T_m is $T_m = 2 \times (A+T) + 4 \times (G + C)$. That is, 2 times the number of As and Ts, plus 4 times the number of Gs and Cs. T_m should be well below 80 degrees C: above this temperature, the polymerase used in PCR degrades!

Thus a really long primer will have a really high T_m , and will be difficult to use.

The entire bacterial genome has about 1,000,000 base pairs. If your primer is 4 base pairs long, what are the odds that it will randomly match a sequence in the genome? (Hint: What is the probability of a specific sequence of 4 base pairs occurring, when each has a 1/4 chance of occurring?) What about a specific sequence of 10 base pairs? How about 100?

What length primers will you choose so that the T_m is low but the primer is still as specific as possible? (There is no one right answer, here, there are several lengths that will work just fine!)

What is the probability that a primer of this length will randomly occur in the bacterial genome?

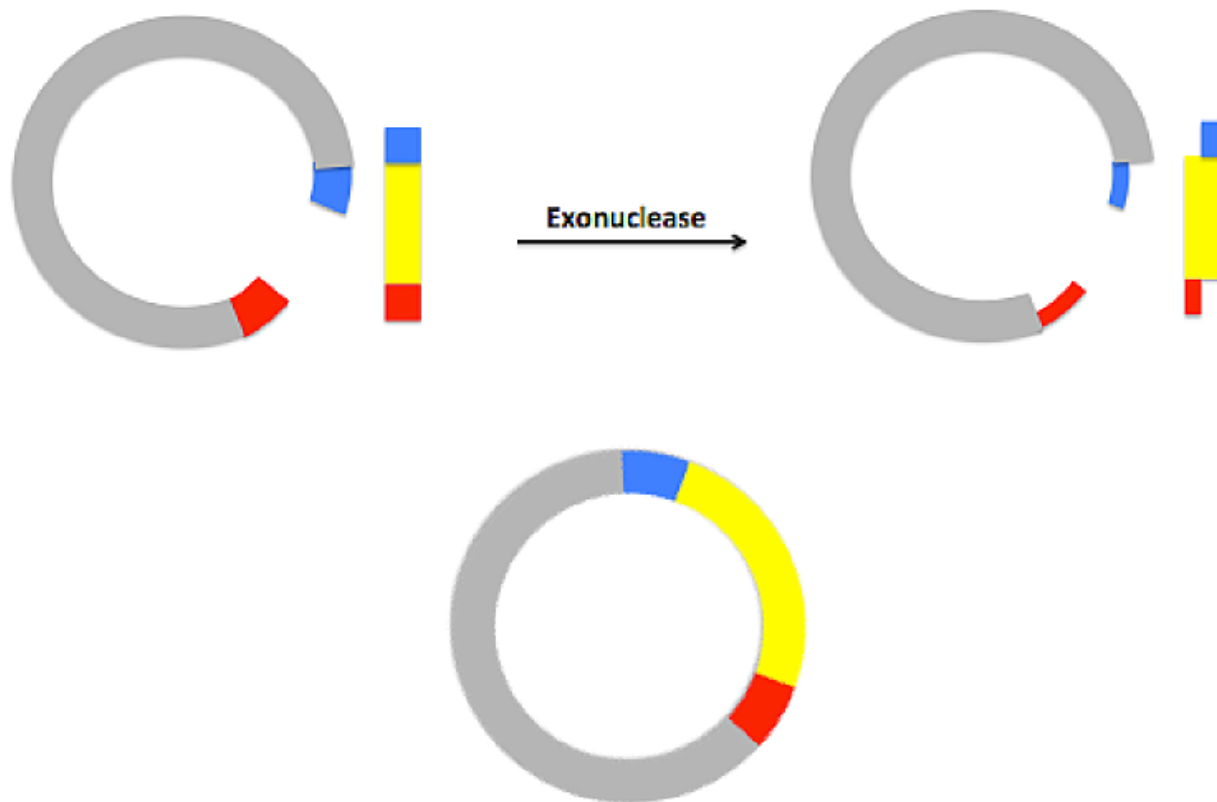
What is the sequence of the primer for the blue strand (it should bind to the last (your choice of length) base pairs of the blue coding strand target region)?

What is the sequence of the primer for the green strand (it should bind to the first (your choice of length) base pairs of green coding strand target region)?

What is T_m for your primers? Use the formula on the previous page to calculate this value. Show your work.

Designing Gibson Primers

Gibson assembly cloning is a recently discovered and popularized method of combining pieces of DNA into a single plasmid.



Gibson assembly cloning is a recently discovered and popularized method of combining pieces of DNA into a single plasmid.

When designing primers, they must be complimentary to either of the two DNA strands on the insert and on the backbone.

Now that you understand how to design primers for PCR, can you design primers to attach these complementary DNA regions to the ends of a backbone, and to the ends of an insert?

Transformation

H Biology

WHAT IS RECOMBINANT DNA?

time: 90 minutes



Learning Objectives

Students will use paper to model recombinant plasmids, in order to understand how plasmid DNA can be used in synthetic biology. Students will add an insert of interest (for example, an insulin producing gene from a mammal) into a bacterial vector. They will review membrane properties and learn how plasmid DNA can enter bacteria through horizontal transfer. They will discuss how scientists select for bacteria that took up the plasmid. Finally, students will examine the costs and benefits of using plasmid DNA instead of chromosomal integration.

Materials

- Use the included Transformation handout to make inserts and backbones
- Computer with Internet access to find DNA sequences for genes of interest
- Tape
- Scissors

Procedure

Cut out backbones and inserts using the included handout. Find a gene of interest online (websites given on the handout) and write the ends of the sequences on the insert. Design or discuss primers that would be necessary in the Gibson assembly of the insert into the backbone. Include an antibiotic resistance gene to allow selection once the bacteria are grown on plates. Go through transformation steps, discussing how each step affects the cell membrane and where the plasmid of interest is at a given time.

Approximate Cost: free with a computer!

Background

Transformation is a process by which scientists can introduce foreign DNA into a bacterial cell. Because bacteria have no nuclear envelope (they are prokaryotes), plasmids transformed into the bacteria will be translated/transcribed in the same way as the chromosomal DNA. The main difference is that while there is only one chromosome, there may be many plasmids! During transformation, some bacteria may pick up only one plasmid, while others may pick up two or three or even a hundred! This can cause some difficulties when using bacteria as vectors for biological devices from which a scientist is trying to take measurements. In the case of insulin producing bacteria, though, this just means some bacteria will make more insulin than others.

For details on Gibson Assembly, please see the handout and this video: <https://www.youtube.com/watch?v=eLJLKyHFqTg>

For details on what each step of a chemical transformation and electroporation does, please see the handout.

Critical Thinking Questions

What would a scientist do to transform a plasmid into a bacterium that already has a plasmid transformed into it?

What are the pros and cons of different types of transformation (chemical vs. electroporation)?

Why is it important to inoculate single colonies within 24 hours? (Think about horizontal gene transfer, mutations, and antibiotic resistance.)

BacteriMeasure

H Biology

WHAT ARE APPLICATIONS OF SYN BIO?

time: several days



Learning Objectives

In this lab, students will build their own biological device, expressed in bacteria. This is an excellent application of synthetic biology, and students will have the opportunity to design primers for recombinant DNA, transform bacteria with the plasmid of interest, grow the bacteria on plates, and visualize lacZ expression based on RFP intensity. This lab introduces the concept of transcriptional noise.

Materials

- There are many materials required for this lab. See the materials list on the included handout.

Procedure

This lab is based on:

http://openwetware.org/wiki/BioBuilding:_Synthetic_Biology_for_Students:_Lab_2

We'll be making a handout with the instructions for our own version, more focused on promoters, transcriptional noise, and E. coli, to include with this curriculum packet soon!

Approximate Cost: \$50 for 25 students

Background

Synthetic biologists are engineers, and therefore engage in the design > build > test process.

Synthetic biologists design and build genetic devices to induce expression of a particular protein or function in an organism.

Each genetic device requires a promoter and ribosomal binding site (RBS), which control transcription, a coding sequence for the protein or function, and a terminator to end transcription after the coding sequence.

It may be necessary to tune the level of output of a particular genetic device. A synthetic biologist doesn't necessarily want a lot of output of a protein he or she is causing the organism to express. One way to tune the level of expression is to change the level of transcription by utilizing a different promoter or RBS. The lacZ coding sequence can be used to measure strength of the promoter and RBS. lacZ codes for expression of beta-galactosidase, which can metabolize ONPG (o-nitrophenyl- β -D-galactoside) into galactose and o-nitrophenol, a yellow compound. More yellow observed when the experimenter gives the bacteria ONPG indicates stronger expression of lacZ and therefore a stronger promoter or RBS.

Critical Thinking Questions

Why might one not always want strong expression of a genetic device? A synthetic biologist needs to be thoughtful of how the genetic device interacts with the rest of the cell's functions. In many cases, it is ideal for the device to be...

To be finished once we have the handout!

if A, then T

H & AP Biology

PROGRAM A REVERSE COMPLEMENT CALCULATOR

time: 45 minutes



Learning Objectives

Biology has rapidly become a big data science in which practitioners have to handle terabytes of data. This can't be done manually, so the ability to process and make sense of large amounts of data is becoming increasingly necessary. (i.e. RNA-Seq, in which the level of expression as measured by RNA transcription for a large array of genes is measured in a cell or population of cells.)

In this module, students will be exposed to basic programming skills, using an online Python simulator.

Materials

- Each student or group of students will need a computer open to:
<http://www.pythontutor.com/visualize.html#mode=edit>.
- It is also helpful to project this page onto a screen in the front of the room for students to follow along if they are having trouble.

Procedure

There are a few basic Python structures students will need to understand to program their reverse complement calculator.

Start by having them write down the steps they take as humans to find the reverse complement of a sequence. Then move onto explaining the structures on the next page. Try each structure (if, else, for, input, print, [::-1]) out on the Python simulation website linked in **Materials**. Then, in groups or as a class on a projector, try coding a reverse complement calculator on the Python simulation website!

Approximate Cost: free with a computer!

Background

Here is one example of interactive working code: <http://tinyurl.com/wmigemRevCalc>
The same code is printed below for your convenience.

//comments are given in blue after the double slash and do not code for anything.

```
print("Welcome to W&M iGEM's Reverse Complement Calculator!") //prints this line in the console. Don't forget quotes!
```

```
sequence = input("What is your sequence? ") //Asks the user for a sequence and sets that sequence as a variable called sequence.
```

```
revSequence = "" //makes a variable, revSequence, to store the reverse complement sequence.
```

```
validSeq = True //a boolean (can only be True or False) variable
```

```
//this for-loop finds the complement of each base
```

```
for base in sequence: //goes letter by letter (base by base) through the given sequence
```

```
    if(base == "A"): //if the current base is A...
```

```
        revSequence += "T" //add T to the revSequence
```

```
    elif(base == "T"): //otherwise if the current base is T...
```

```
        revSequence += "A" //add A to the revSequence
```

```
    elif(base == "G"): //otherwise if the current base is G...
```

```
        revSequence += "C" //add C to the revSequence
```

```
    elif(base == "C"): //otherwise if the current base is C
```

```
        revSequence += "G" //add G to the revSequence
```

```
else: //if the current base isn't A, T, C, or G...
```

```
    print("Invalid base: "+base) //prints "Invalid base: " and the incorrect base in the sequence the user gave
```

```
    validSeq = False //if the current base isn't A, T, C, or G the sequence isn't valid anymore
```

```
    break; //stops going base by base, because the sequence isn't valid
```

```
revSequence = revSequence[::-1] //reverses revSequence (puts the characters in backwards order, so that we now have the reverse complement.
```

```
if (validSeq): // if the sequence was valid, validSeq will still be True, and this if statement will continue.
```

```
    print("The reverse complement of "+sequence+" is "+revSequence+". Thanks for coding with us!")
```

```
else: //if validSeq is not True, aka validSeq is false (if there was a base that wasn't A, T, C, or G)
```

```
    print("Sorry, you didn't provide a valid sequence.")
```


RFP Art

H & AP Biology

HOW ARE FLUORESCENT PROTEINS USED?

time: 45 minutes



Learning Objectives

Students streak a plate, a protocol commonly used by synthetic biologists to grow up cells with modified DNA. Students should understand that cell division, and therefore bacterial growth, is exponential. Students should also understand that a colony grows from a single cell and that all cells in a colony have identical DNA unless the DNA of a cell mutated.

Materials

- Bacteria with an RFP (red fluorescent protein) gene <http://www.enasco.com/product/LM00724M>
- Agar Plates (one per pair) <http://www.enasco.com/product/Z13903M>
- Toothpicks (one per student)
- Permanent Marker

Procedure

Explain the idea of plasmids to students. Ask them what type of protein they would design a plasmid to code for if they were trying to see whether or not there was a plasmid in a cell. Introduce them to the idea of using a colored protein as an 'indicator' that signals to the researcher whether or not inserting a plasmid into a cell was successful. Ask students how cells grow: if a cell has an RFP plasmid in it, will it grow into a red colony? At what rate do cells grow? If one cell becomes two cells in twenty minutes, then how many cells will you have in twenty minutes if you have eight cells now?

Give each pair of students an agar plate and have them write their names on the lid of the plate. Turn the plate upside down and draw a line across the bottom, dividing the plate in half. Give each student a toothpick. Each student should take turns dipping the toothpick into the bacterial culture and then streaking half of the plate with it in any pattern they want. Don't press too hard with the toothpicks - they shouldn't break the surface of the agar. Store plates at 37 degrees Celsius overnight or at room temperature for 48 hours. Students can take pictures of their plates and see if their designs grew.

Approximate Cost: \$25 for 20 students

Background

A plasmid is a circular molecule of DNA that a bacteria can take up and express. For molecular biology, a plasmid consists of a vector backbone, which carries a particular antibiotic resistance gene, and the insert, which is some genetic device or component of a device.

RFP is red fluorescent protein, which fluoresces red under certain wavelengths of light.

In epifluorescent microscopy, a laser is used to illuminate an organism expressing a fluorescent protein such as RFP. Computer software that works with the microscope can detect the level of fluorescence, which tells us how strongly the fluorescent protein is expressed.

Fluorescent proteins like RFP are used as reporters in synthetic biology, sometimes just to indicate that a device works, and sometimes to signal some event, where activation of RFP is the last of a series of steps in a genetic device's function.

Example: A biosensor is an organism programmed to detect a particular substance in the environment. Here is an example of a device designed to detect cobalt, a metal that can be toxic to humans:

http://parts.igem.org/Part:BBa_M45102. The promoter, which controls gene transcription, is sensitive to cobalt. Cobalt in environment of a bacteria expressing this device will have the gene activated by cobalt and express RFP, which serves to report the presence of cobalt.

Critical Thinking Questions

Looking back at the structure of plasmids, notice the backbone contains antibiotic resistance? Why would this be?

In a biosensor like that described above, what would levels of intensity of the fluorescence indicate?

Plasmid Design

AP Biology

SOLVE A PROBLEM WITH SYNTHETIC BIOLOGY

time: 60 minutes



Learning Objectives

Students will learn what plasmids and genes are, and they will design a plasmid that could be synthesized and put into a bacterial cell. This gives students an insight into the planning that synthetic biologists do every day. It also introduces them to the idea that all engineers, including genetic engineers, design then build then test all their projects.

Materials

- One Plasmid Design worksheet per student (see template)
- Optional: computer to look up sequences on the iGEM Registry

Procedure

After discussing what a plasmid is with students, students should complete the provided worksheet. There are many resources students can use to look for part sequences, but we recommend the iGEM registry (a Google search will direct you there). On the registry, parts, like a promoter region, ribosome binding site, double terminator, and the student's gene of interest, can be searched by function or by name. Allowing students to browse this site, explore other teams' past projects, and sorting through part descriptions, will give students a sense of what many synthetic biologists go through in the process of designing a project. Ask students for a problem they want their plasmid to solve. For example, one student may want to use bacteria to produce insulin. That student would then choose an appropriate promoter, RBS, and double terminator, and use the part sequence for part BBa_K1328003, a human insulin encoding gene, as the gene of interest.

Approximate Cost: \$8 for 25 students

Background

See the provided handout with all necessary background information. Also see the iGEM registry for information on different genes of interest, in case you would like to select some genes in advance for students to choose from or put together (i.e. to make bacteria turn red and to produce a particular toxin).

Critical Learning Questions

What protocols would you use to put together these pieces of DNA into a plasmid, and then to put that plasmid into bacteria?

Plasmid design is an example of synthetic biology. What sorts of problems do you think could be solved using synthetic biology? How? Search online to see if any scientists are currently researching your idea!

What is a plasmid?



This is a plasmid.

It is a small, circular piece of DNA that scientists can easily put into bacteria. It is completely separate from the genome, the main piece of DNA in a cell. If the genome is like a book, a plasmid is a postcard.

The two main parts of a plasmid are:

1. the vector (grey)

The vector is a vehicle for the construct. A vector is a blank postcard: it has a purpose and can be sent into the cell, but there is no writing on it, so it can't communicate anything to the cell on its own.

2. the construct (colored)

The construct includes one, sometimes more, gene. A single gene typically codes for a single protein. A construct is writing on a postcard. It communicates a message to the cell, but it needs to be a postcard to do so.

This is a gene.



It codes, or has instructions, for one complete protein. A construct is made of one or more genes. Each gene is like a sentence; it has a start, a message it communicates, and an end.

The four main parts of a gene are:

1. the promoter (blue)

This is the start of a gene. It tells the cell to begin reading the gene by providing a place for RNA polymerase to bind to the DNA. It acts like a capital letter at the beginning of a sentence.

2. the RBS: ribosome binding site (pink)

This is the second half of the start of the gene. It tells the cell to start understanding the gene by providing a place for a ribosome to bind. It acts like the first word in a sentence.

3. the coding region (yellow)

This is the main part of the gene. It is a DNA sequence that contains the instructions for synthesizing a protein. The coding region acts like the middle of the sentence.

4. the terminator (navy)

This is the end of the gene. It tells the RNA polymerase to fall off of the gene and stop reading the plasmid. The terminator acts like the period at the end of a sentence.

Design a plasmid

Scientists who design plasmids and put those plasmids into cells are called **Synthetic Biologists**. Now it's your turn to be a synthetic biologist!

Manual PCR

AP Biology

HOW DOES A THERMAL CYCLER AMPLIFY DNA?

time: 180 minutes



Learning Objectives

In this module, students will replace the thermal cycler and amplify DNA manually, using several hot water baths. They will examine how different temperatures interact with the DNA molecules and primers, and how these changing temperatures can be used to conduct a polymerase chain reaction by hand.

This module should reinforce base pairing rules, understanding of DNA strand directionality, and the process of DNA replication.

Materials

- DNA Sample
- Primers
- PCR beads
- Pipettes
- Distilled water
- Thermometers
- Hot water/hot plates
- Several Tupperware containers
- Tongs
- Endurance

Procedure

This activity requires two water baths: one at 55 degrees C and the other at 98 degrees C. A hot plate may be used to maintain these temperatures, or water can be continually microwaved and switched out. Combine 1 uL sample DNA, 2 uL of each 10 uM primer, distilled water, and PCR beads.

First, and *only once*, hold the tube in 98 C for 30 seconds. Then, begin cycling as follows. Do this cycle 30 times.

- 98 degrees C for 10 sec
- 55 degrees C for 30 sec
- 72 degrees C for 60 sec
- 72 degrees C for 2 min

Approximate Cost: \$125 for 25 students

Background

Polymerase chain reaction (PCR) is used routinely in biology labs around the world. The PCR reaction allows scientists to amplify a specified region of the DNA using specially designed primers that scientists order themselves. For more information on the primers, please see the **Primer Design** activity.

Each step of PCR changes the sample temperature, which affects the enzymes within the PCR bead. A review of the steps in DNA replication should help students understand how we use a polymerase inside the PCR bead to replicate DNA for us. A review of environmental factors on protein structure (see Egg Science) should help students understand how different temperatures can affect primer binding.

Detailed information on how each step of PCR works can be found at: http://www.abpischools.org.uk/page/modules/pcr/pcr2a.cfm?coSiteNavigation_allTopic=1

Critical Thinking Questions

What protocol do scientists use to see if their PCR amplification was successful?

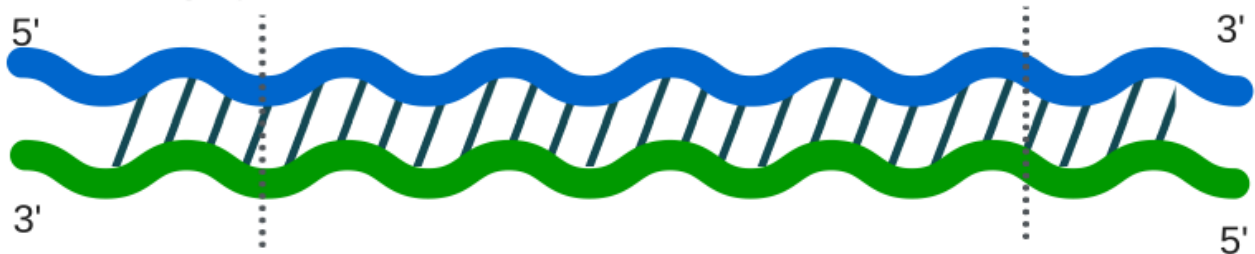
If two bands appear on a gel after gel electrophoresis, which is used to check if PCR amplification was successful, and only one band is correct, what is the next step in optimizing PCR for your sample and primers?

If the T_m (melting temperature) of the primers is 62 degrees C, what temperature should the annealing step of PCR be?

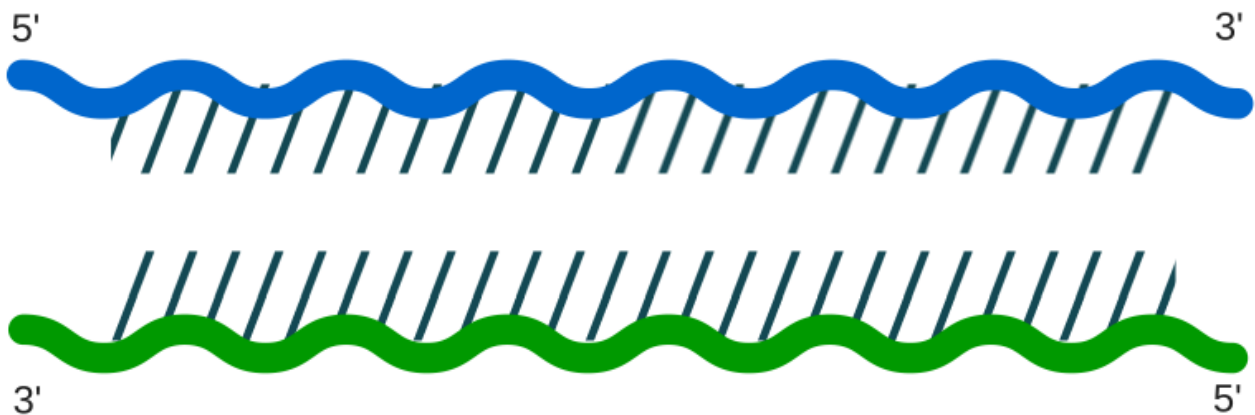
If the strand one is trying to amplify is longer or shorter, what step in the cycle should be adjusted?

The PCR Process

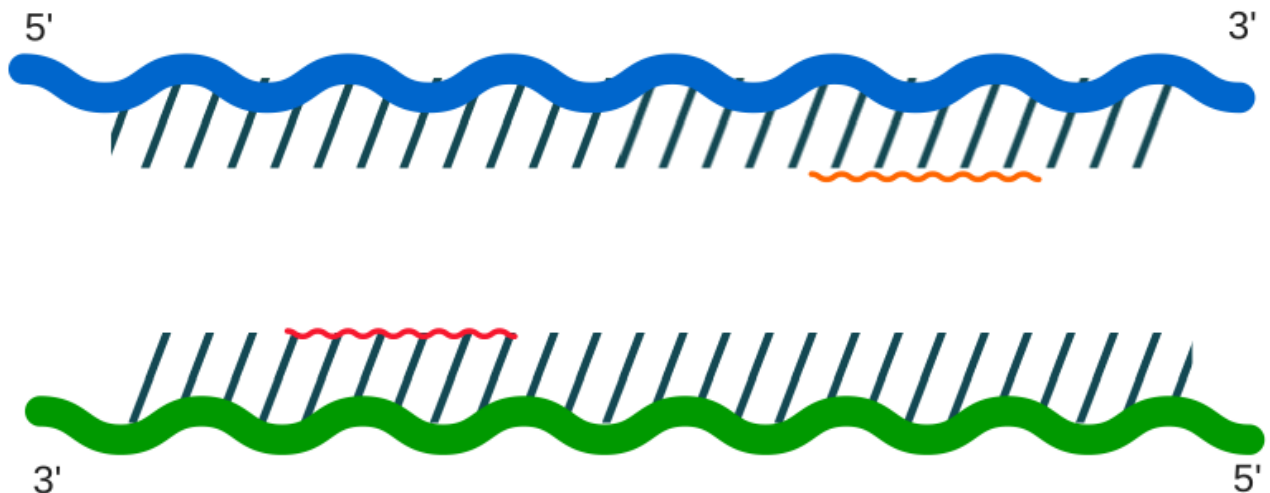
Double stranded DNA sequence. The grey lines show the region that we want to amplify:



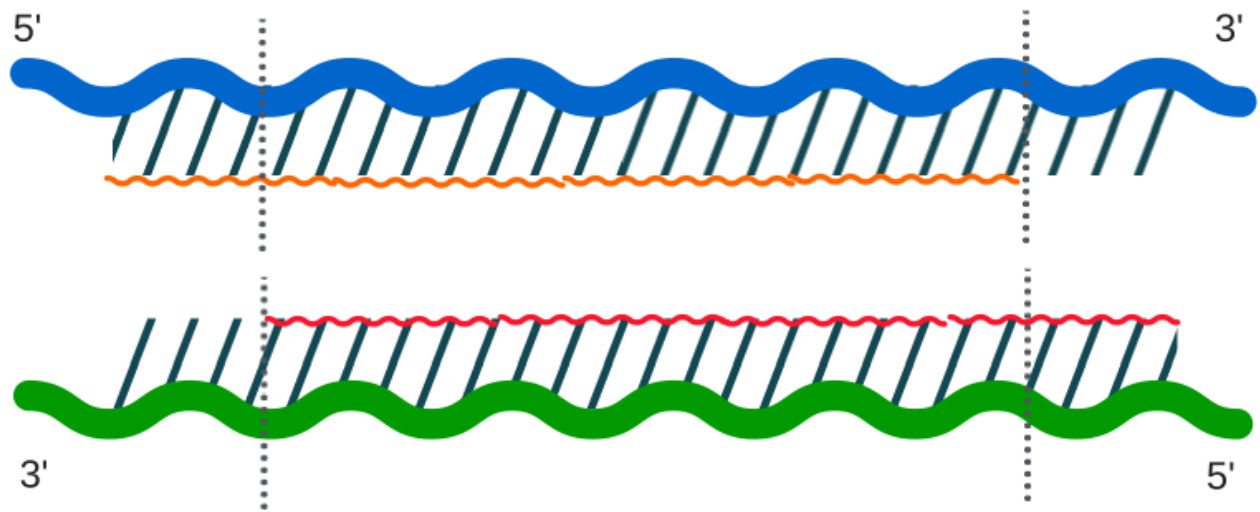
Melting step disrupts the hydrogen bonds linking base pairs together:



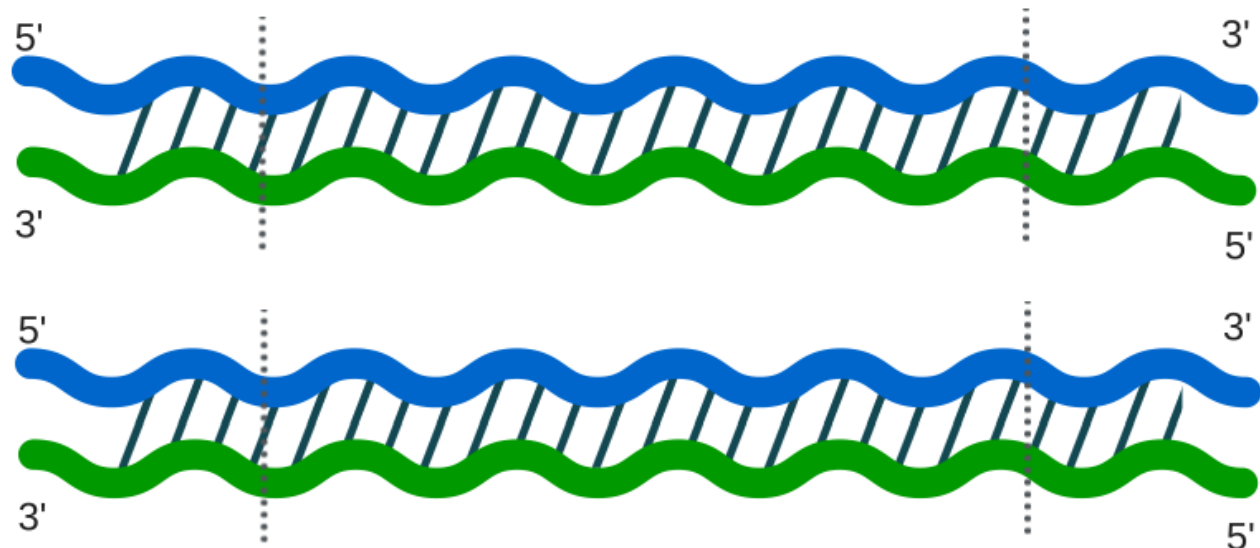
A lower temperature annealing step allows forward and reverse primers to bind (this is quicker than the time it takes template strands to come back together). Primers bind to the template where their sequences match up.



In the extension step, we raise the temperature again so that the polymerase can extend the primers by adding nucleotides to the 3' ends.



Now we have two copies of the region of interest!



Now we have doubled the number of template strands. These two strands will start again at the beginning of the cycle, split during the melting step, bind to primers at the annealing step, and form four new copies of our double stranded DNA. It is in this way that DNA can be amplified.

SynBio Ethics

AP Biology

WHAT ETHICAL CONCERNS SURROUND SYN BIO?

time: 60 minutes



Learning Objectives

Students should consider the connection between scientists and the public. They should recognize some of the many ethical concerns that arise when new technologies are implemented in scientific research, especially in biological research. They should question the responsibility scientists have for their research.

Materials

- A computer or printouts of "Ethical Issues in Synthetic Biology" found at www.synbioproject.org/process/assets/files/6334/synbio3.pdf

Procedure

Before discussion:

Introduce the concept of synthetic biology.

Read and have students read "Ethical Issues in Synthetic Biology" before class, either as homework or as a group prior to the discussion. Either print a few copies of "Ethical Issues", let students pull it up on computers, or project it at the front of the room, so that students can follow along during the discussion.

Discussion:

Ask students to summarize the main points made in "Ethical Issues". Ask students what their concerns about synthetic biology are, and keep a list of concerns. Then, ask students to split into small groups. Each small group should assume the role of a synthetic biology researchers and come up with strategies to address three of the concerns on the list. After 15 minutes, discuss the strategies students came up with as a class. Do real scientists follow these strategies? How does research funding affect scientists' ethical decisions? Is it morally permissible for scientists to research new methods and biotechnologies without considering the impact their research might have on the public?

Approximate Cost: free!

Background

According to SyntheticBiology.org: "Synthetic biology is a) the design and construction of new biological parts, devices and systems and b) the re-design of existing natural biological systems for useful purposes". As such, synthetic biology is a cross between biology and engineering. Synthetic Biology is a relatively new field, less than twenty years old, and like all new scientific fields it has many ethical questions that have yet to be answered by the field as a whole. Students have the unique position of receiving a scientific education and being able to address the ethical questions surrounding the field with fresh eyes, and they can address the questions from the view of the public and the view of researchers.

Before leading this discussion, it is important to ensure that students will approach the discussion using logic and addressing the strengths and flaws of arguments made rather than judging the students who present those arguments. The topic of the ethics of synthetic biology, like any ethics topic, can be a sensitive subject. However, the skill of evaluating the goals, benefits, and disadvantages of research is invaluable to both scientists and the public, and as such should be developed early.

Critical Thinking Questions

Historically, how have societies overcome controversies about new, rapidly developing technologies?

Do synthetic biologists who are modifying organisms that are meant for human consumption have a larger responsibility for their research than most scientists? Why or why not?

What are the benefits of genetically modified organisms (GMOs)?

What are the potential negative effects of GMOs?

Give it a try!

W&M iGEM

SYNTHETIC BIOLOGY CURRICULUM

Special thanks to Ms. Bonnie Ellis, Science Curriculum Coordinator for Williamsburg-James City County Public Schools, for helping us share this curriculum!

If you are a teacher in the Williamsburg area, members of our team are happy to come to your school and help run these activities this fall. E-mail igem@wm.edu to schedule!