Rieder - Genetics **Reading #: _____**
Worksheet 10: **Biotechnology!**
Due: 12/12/17 at the beginning of class

These will be returned in my faculty mailbox by Friday 12/15.

Name: _____

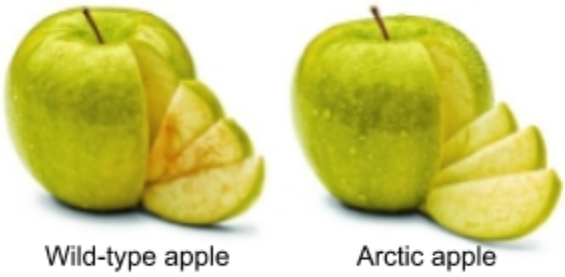
About how long did this homework take you? _____

I consulted/worked with: _____

I will not accept late homework. Special exceptions will be made **only** in the event of illness or if you contact me at least 24 hours ahead of when the assignment is due (at my discretion).

POINTS: / 50

Arctic® apples recently went on sale at select markets in the Midwest. These apples are **genetically engineered** (a synonym for **genetically modified**). Read:



"How'd we 'make' a non-browning apple?"

(https://www.okspecialtyfruits.com/howd-we-make-a-nonbrowning-apple/)

1. How many apple genes contribute to the production of polyphenol oxidase (PPO) (2 pts)?

2. What is the **phenotype** of an apple that **expresses** all the genes that contribute to PPO production (1 pt)?

3. What is the special **phenotype** of an Arctic® apple (1 pt)?

Scientists have introduced **transgenes** into the apple genome to make the Arctic® apple. Please read about this introduction "How we introduce the nonbrowning trait in Arctic® apples" (https://www.okspecialtyfruits.com/how-we-introduce-the-nonbrowning-trait-in-arctic-apples/)

4. What organism do scientists use to get the **transgenes** into the genome of the Arctic® apple (3 pts)?

You may notice from the above reading that the scientists are genetically modifying leaf cells... But you learned (from your Thanksgiving homework question on sweet potatoes) that modifying leaf cells will NOT modify all the cells in the plant!

5. Are leaf cells **totipotent** or have they **differentiated** (2 pts)?

Scientists can grow a ball of leaf cells in dishes in the laboratory-- this is called a callus.

To make a **genetically modified** apple, scientists first genetically modify calli cells.

Second, they **de-differentiate** the leaf cells so that they are now **totipotent**.

The calli can now grow into whole trees!

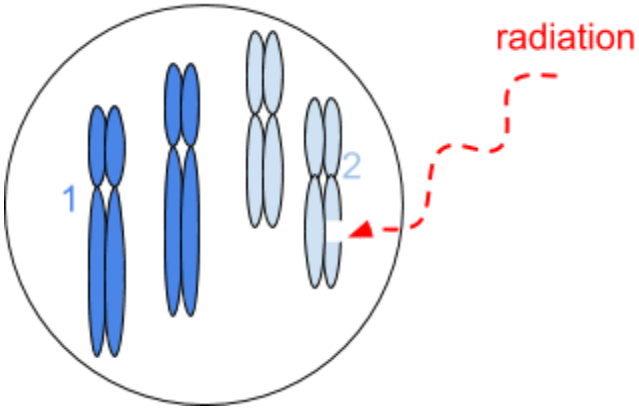

Calli (many calluses)

The discovery that you could de-differentiate cells and give them *back* the property of totipotency (usually reserved for embryonic cells) was the subject of the 2012 Nobel Prize for Physiology or Medicine, given to Sir John B. Gurdon and Dr. Shinya Yamanaka. (https://www.nobelprize.org/nobel_prizes/medicine/laureates/2012/press.html)

6. Once scientists have put the **transgenes** into a single Arctic® apple tree, how do they propagate the tree (1 pt)?

7. The articles you have read so far about the Arctic® apple discuss a technique called "gene silencing." This is another name for **RNA interference** (**RNAi**). What Arctic® apple gene(s) are the targets of RNAi/gene silencing (2 pts)? Please do not write "nonbrowning genes," but be slightly more specific.

8. The cell pictured below is undergoing ***meiosis*** and has already replicated its DNA. But one **sister chromatid** of chromosome 2 has suffered DNA damage due to radiation (like sunlight, for example). This chromatid has experienced a **double-stranded DNA break**! The cell now completes **meiosis** without repairing this damage. Please draw the remaining stages of **meiosis** and the resulting **gametes** (6 pts):

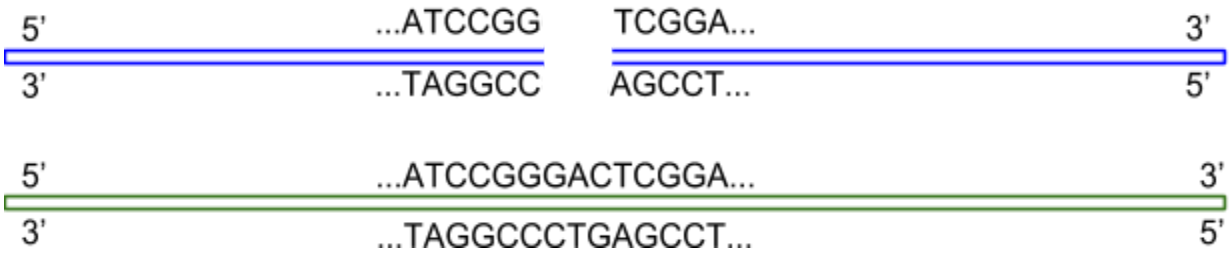


9. How many of the gametes you made in question 8 can undergo **fertilization** to make viable embryos (1 pt)?

_____

When a cell--either **somatic** or **germ**--suffers a double-stranded DNA break, its number one objective is to repair that DNA! In class we learned about the two mechanisms of **DNA repair**: **Homologous Recombination** (**HR**) and **Non-Homologous End Joining** (**NHEJ**).

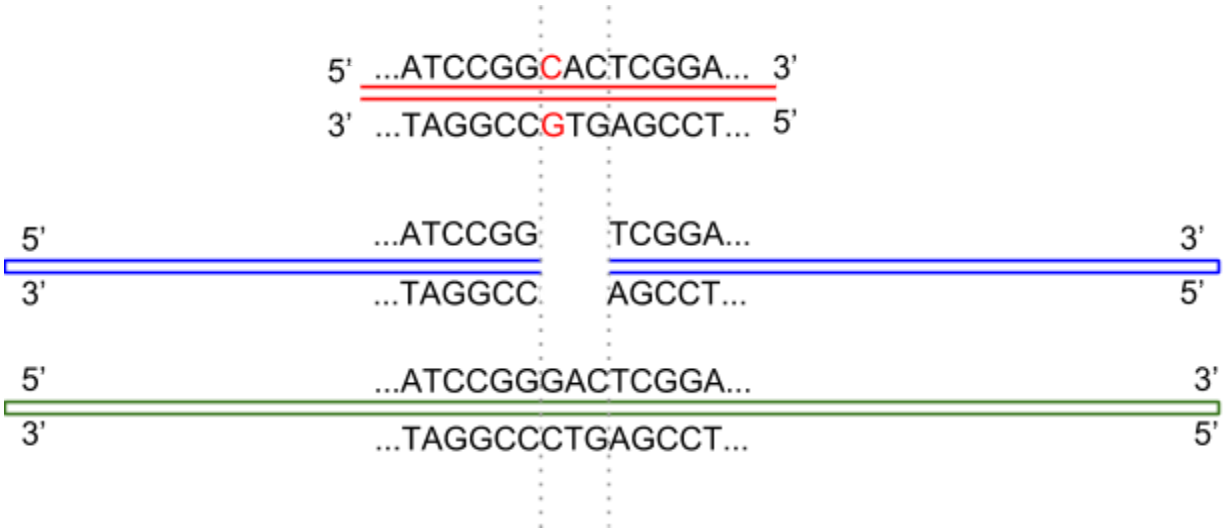
Below is a **double-stranded DNA break** that the cell will repair. These are ***sister chromatids***. The sequence around the breakpoint is noted.



10. The cell repairs the above **double-stranded DNA break** using **HR**. Is it likely that this will result in a mutation in the blue chromosome? If so, what kind of mutation do you predict (3 pts)?

11. The cell repairs the above **double-stranded DNA break** using **NHEJ**. Is it likely that this will result in a mutation in the blue chromosome? If so, what kind of mutation do you predict (3 pts)?

12. A cell that is not dividing experiences a **double-stranded DNA break**. Which DNA repair mechanism (**HR** or **NHEJ**) is the cell more likely to use (2 pts)?



Above is the same **double-stranded DNA break**. However, now a scientist has injected a small fragment (red) of double-stranded DNA into the cell. The fragment can act as a repair template. The cell will repair this dsDNA DNA break using either HR or NHEJ.

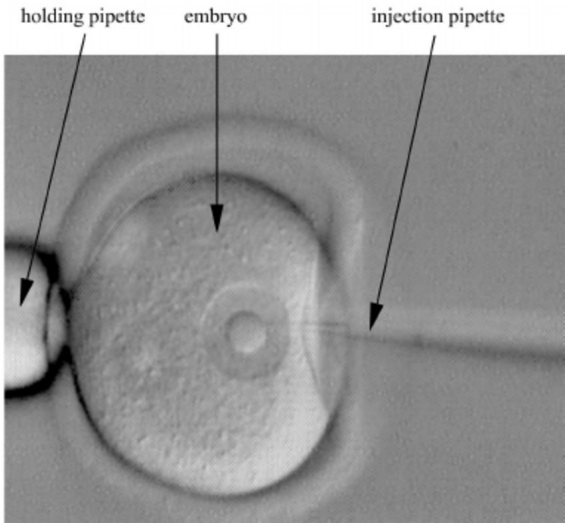
13. Draw the stages of **HR** (strand invasion, synthesis, and resolution) using the repair template (6 pts).

You cannot control where in the genome radiation (like UV or X-rays) will cause a double-stranded DNA break. So if we want to introduce a change (mutation) into a certain gene, the most difficult part is to target the double-stranded DNA break.

14. Below, fill out the ***three*** strategies that scientists use to make **targeted double-stranded DNA breaks** (6 pts).

Strategy name	Need to make a new protein (Y/N)?	"Off target" effects (many/few)?	How easy to engineer (easy/difficult)?

You have a single-celled mouse **embryo** (below). This embryo has a **wild-type** genome, but you want to use it to study the human disease Amyotrophic Lateral Sclerosis (ALS). To do this, you must engineer the embryo's genome to include a **missense mutation** in the *SOD1* gene, which leads to ALS in humans.



15. You want to use CRISPR to introduce the *SOD1* missense mutation into the mouse genome. What do you inject into the embryo (3 pts)? Be specific, please.

16. You forgot to inject the repair template! What are two possible outcomes? In other words, what will happen to the embryo's *SOD1* gene? What are possible mutations (4 pts)?

1.

2.

17. Take the NY Times quiz: "Can Gene Editing Actually Do That?" (https://www.nytimes.com/interactive/2017/08/04/science/crispr-gene-editing.html?_r=0)

Describe something from this quiz that the CRISPR/Cas9 gene editing technology has been used for (click on one of the hyperlinks from a question you find interesting). How is this particular application of the CRISPR/Cas9 technology useful (4 pts)?

In our very last class of the semester (12/12) we will have a class-wide discussion about **genetically modified organisms** (**GMOs**) (also called **transgenics** by scientists). For participating in this discussion you can ***earn up to 3 extra points on your final exam grade***.

I will keep track of who is participating. You will earn a point (at my discretion) each time you make a comment that is:

1) Relevant and substantial
2) Not something already said
3) Based on what we have learned in class or from the assigned readings

For better or worse, GMOs are controversial. GMO foods are sometimes even called "Frankenfoods" and a quick google image search for "GMO" turns up some plainly ridiculous images meant to scare people. Sometimes this is because people don't understand genetics, but sometimes people raise valid concerns. It's *especially* important to be able to distinguish valid points from those formed from a misunderstanding of genetics.

Why is it that G.M.O.s, more than any other food issue, have inspired so much angst? "There's something about genes that just terrifies people, when, in fact, this method is just as safe as the plant breeding we've been doing for ten thousand years," [Pam] Ronald said. Grist.org's [Nathanael] Johnson told me that people see genetic engineering as "a form of tinkering with the very essence of the life force, so it lends itself to all sorts of ominous metaphors."

- Amanda Little, "A Journalist and a Scientist Break Ground in the G.M.O. Debate," *The New Yorker*

Everybody read the article: "U.S.D.A. Approves Modified Potato. Next Up: French Fry Fans." (NYTimes).

Would YOU choose to eat French fries or mashed potatoes made from this new GMO potato? Why or why not?

You may read your reading or multiple readings (as long as one of them is the one assigned).

Reading #1: "A Race to Save the Orange by Altering Its DNA" (NYTimes).

Reading #2: "In Midwest, Flutters May Be Far Fewer" (NYTimes).

Reading #3: "Genetically Engineered Salmon Approved for Consumption" (NYTimes).

Reading #4: "Farmers Cope With Roundup-Resistant Weeds" (NYTimes).