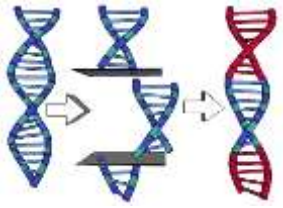


Name: _____ Date:_____

Gen Bio 1 Lab #11: PCR Gel & GATACCA

<u>Pre-Lab Reading</u>: Pages 331-339 9th Ed. Complete Pre-Lab vocabulary and Pre-Lab Questions before attending Lab.

<u>Pre-Lab Vocabulary:</u>

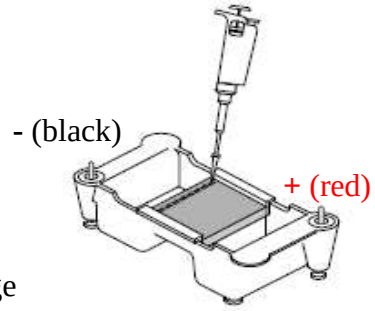
1. Agarose gel electrophoresis –
2. DNA molecular weight ruler –
3. QUIKView DNA stain –
4. Genetic engineering –
5. Genetically modified crops –
6. Genetically modified food controversies –

Procedure 1: Analyzing our PCR results using Agarose Gel Electrophoresis

Today we will run out our PCR samples from last week, hopefully observing strong detection bands of our amplified DNA. Each group will be pouring a gel, and running their samples independently. Note that we all need to wear gloves today, because the chemical we use to stain our agarose gels is toxic.

Materials

Gel casting tray with dams and a comb
Electrophoresis chamber
Running buffer (TAE 1X concentration)
2.0% agarose (premixed)
Eppendorf Micropipette
Pipet tips (100 or 200)
From Bio-Rad freezer kit: PCR molecular weight ruler, 200 µl & Orange G loading dye, 1ml
Plastic 1ml pipettes
PCR tubes from last week (stored in the freezer)
Warm or room temp QUIKView DNA Stain (100mL per group)
Staining trays
100 or 200mL Graduated cylinder



Remember: "run to red"

Camera phone (student-provided; photo documentation)

Procedure A: Preparing and loading the gel

1. Prepare gel casting tray and add 30 ml of 2.0% agarose after melting in the microwave. Allow gel to solidify in the refrigerator.
2. Carefully remove the comb from the casting tray and then carefully remove the dams.
3. Place the tray with the gel into the electrophoresis chamber with the wells closest to the **negative (black) electrode** (see **diagram**). Next add running buffer until the top of the gel is covered.
4. Obtain your PCR tube from instructor and pulse-spin the tube for ~3 seconds in microcetrifuge.
5. Using a fresh tip each time, add 10 µl of Orange G loading dye (LD) to each sample and mix well
6. Using a fresh tip each time, load 20 µl of the molecular weight ruler and 20 µl each sample onto your gel in the following order:

Sample	**Lane on gel:**	**Pipette into well:**
PCR tube 1: Non-GMO food control with plant primers	1	20 µl
PCR tube 2: Non-GMO food control with GMO primers	2	20 µl
PCR tube 3: Test food with plant primers	3	20 µl
PCR tube 4: Test food with GMO primers	4	20 µl
PCR tube 5: GMO positive DNA with plant primers	5	20 µl
PCR tube 6: GMO positive DNA with GMO primers	6	20 µl
Molecular weight ruler (NOT into your PCR tubes; onto gel!)	7	20 µl
Leave empty	8	-----

Procedure B: Running the gel

7. Place lid on the electrophoresis chamber plug cord into the power supply, adjust the voltage to **110V** and turn the power supply ON. The **gel will need to run for about 45 min**.
8. Allow samples to run until the "fastest" piece (dye) has moved along about **2/3 of the gel**, and then stop the power supply.

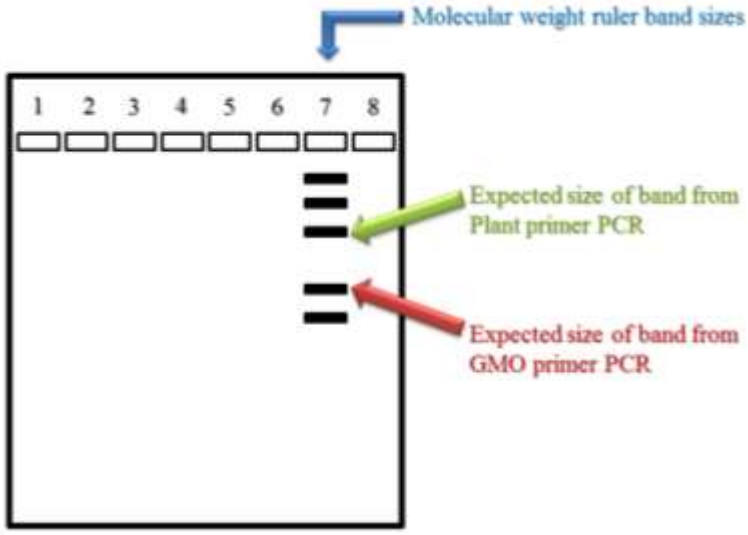
Procedure C: Using QUIKView DNA Stain

9. Gently slide the gel from the casting tray into the **staining tray** and pour approximately about **100 mL of warm dilute stain** into the staining tray so that it just covers the gel.
10. Let the gel stain for **20 minutes**. Make sure the gel remains flat and does not move up against the sides.
11. When finished staining, decant it (pour carefully, while holding the gel gently with your gloved fingers) into a sealable container. **Reduce, reuse, recycle!**
12. Flush the gel under a **gentle stream of tap water**, not directly on gel**, instead on the plastic tray beside the gel**. Do this until the water runs clear.

13. Add distilled water to the staining tray (grey tap) and set off to the side of the sink.
14. After **5 minutes**, carefully pour off water holding onto the gel with gloved fingers.
15. Repeat steps 13 & 14 **four (4) more times**.
16. **View your gel on the light box**. Take an image of this with your phone's camera.
17. Accurately sketch the bands you see on the blank gel in **Question 1** below. Be as exact as possible in sketching the bands in their actual positions. If you choose to take an image, send it to your email, copy it into Word and then print it out.

Questions to be answered by your lab group:

1) **Label your gel picture. Include what sample was loaded into each lane. (You can also attach a printed image of your gel if your group uses a camera phone to capture an image.)**



2) **Which lanes were our "Controls"? What do these controls mean?**

3) **Did your test food contain GMO genes? How do you know?**

4) **Summarize your thoughts on buying foods from the grocery store that contain GMO genes.**

Procedure 2: GATTACA

Instructors: Start movie after all groups have started Procedure 1B

As you watch the movie GATTACA answer the questions below. As we view parts or the entire movie, depending on time, answer the discussion questions.

1) What does Jerome (Vincent) place on the comb at his workstation?

2) "They used to say that a child conceived in love has a greater chance of…." What?

3) What is Jerome's (Vincent's) life expectancy?

4) After Marie's fertilized embryos are screened, how many healthy ones are left?

5) According to the geneticist, we have enough of this built in already. What is it?

6) What is the name given to discriminating against people because of their genetic profile?

7) "After all there is no gene for …" what?

8) What is a "borrowed ladder" or "de-generate"?

9) What does Jerome (Vincent) leave behind at the murder scene?

10) The director claims that Gattaca is occasionally forced to accept candidates with "minor shortcomings", but nothing that would prevent them from working in what field?

11) When Jerome (Vincent) and Irene go to a concert, what is unusual about the piano player?

12) Who killed the mission director?

13) Who does the detective leading the murder investigation turn out to be?

Questions to e x p a n d your mind.

1. As a scientist, you perform the PCR process routinely in your lab. You don't give the process much thought and take it for granted that it works. Recently, a friend without a science background has asked you about the process. Create an analogy to explain an aspect of the PCR process to a nonscientist.

2. What does the movie GATTACA say about DNA determining a person's potential? What are the positive and negative aspects of the world showed in the movie? Provide examples from the movie to support your answer.

3. Examine the two gel results below and determine which baby might have been fathered by Mr. X. Explain your reasoning.