Name: _____ Period: _____

Cellular Respiration Virtual Lab

Note: 1) for your instructor's convenience, please put your answers in blue or red. 2) This virtual lab is based on a video that you can view here (but work through the 1st page 1st)

GOAL: measure and compare respiration rates of dormant and germinating seeds in cold and warm environments.

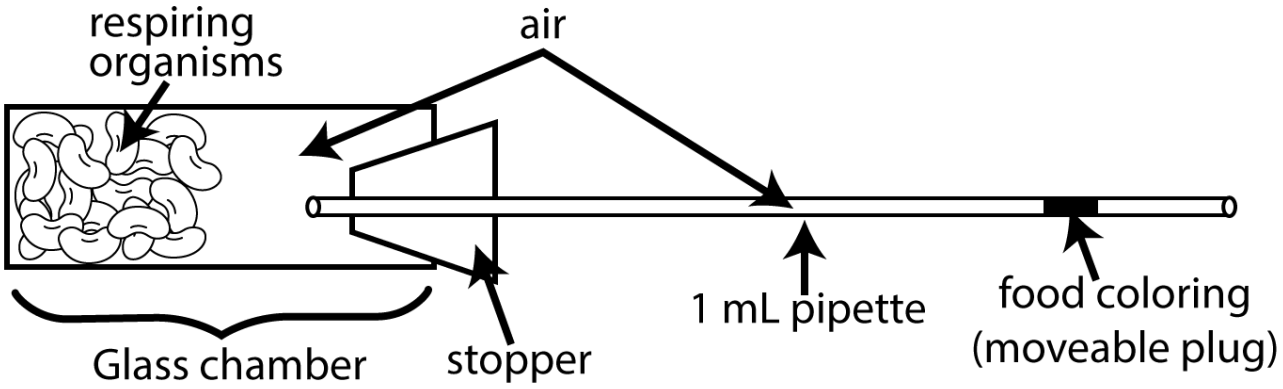
Cellular Respiration:

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + ENERGY$$

How could we measure this?

In this lab, we look at _____. We do this *volumetrically*, using a respirometer

Respirometer design # 1:



Chamber with air, which contains respiring organisms (such as seeds). The tip of the tube is open, so water can enter.

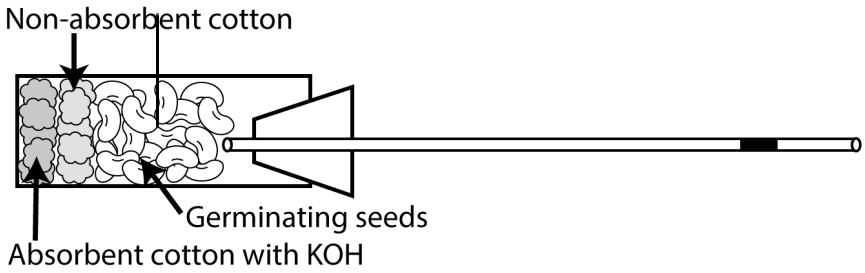
Thought experiment: Assume that the seeds in the respirometer above are respiring. Complete the following sentences

a) The O_2 content of air in the chamber would _____

b) The CO_2 content of air in the chamber would _____

c) What would happen to the volume of air inside chamber and tube? _____

Respirometer design # 2: To see how much O_2 is removed by respiration you have to remove the CO_2 that is produced by cellular respiration: We do this by modifying our set-up as follows:



We soak the absorbent cotton with KOH (potassium hydroxide). KOH reacts with CO_2 as follows:

$$2KOH + CO_2 \rightarrow K_2CO_3 + H_2O$$

K_2CO_3 (potassium carbonate) is a solid precipitate (not a gas) so it has very little volume. Liquid water also has very little volume. What we've done is soak up all the CO_2 produced by respiration.
Make a prediction: *What happens now?* Complete the sentence below

As respiration proceeds_____
_____

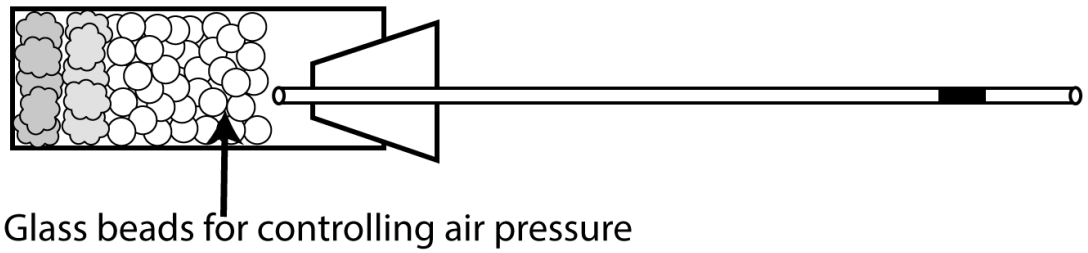
Watch Respiration and Respirometers. Stop at 1:56 (one minutes, 56 seconds)

STILL ONE PROBLEM: In chemistry, you learned the formula PV=nRT (where P = pressure, V = volume, n = # of molecules of gas, R is the gas constant, and T is temperature)? Gas volume is affected by temperature, and temperature is one variable in this experiment.

For example, if we put our respirometer in cold water, what would happen to the volume of gas in the chamber? What would happen to our food coloring plug?

The volume in the gas would _____ The position of the food coloring plug would move _____ because ……..

WHAT CAN WE DO?
Solution: *Another* respirometer w/ glass or plastic beads that we use to monitor pressure changes.

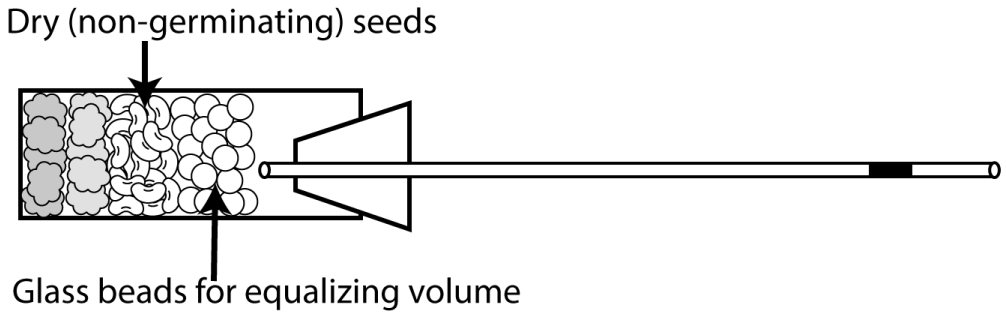


Difference = (initial reading at time 0) – (reading at time X)
Corrected difference = difference from respiration (column b) – difference from beads (column d)

SAMPLE DATA (to demonstrate how to determine the corrected difference)

Time (min)	**Beads**		**Germinating Beans at 20℃**		
	Reading (a)	Difference (b)	Reading (c)	Difference (d)	Corrected Difference
Initial	0.93	----------	0.91	----------	----------
0-5	0.91	0.02	0.84	.07	.05
0-10	0.90	0.03	0.77		
0-15	0.90		0.71		
0-20	0.90		0.64		

Finally, if we want to compare respiration in dry seeds and germinating seeds, we need to account for the fact that the volume of a single germinating seed (which has been soaked in water) will (because of osmosis) be much greater than the volume of a dry seed. If we want to do a one to one comparison, we need to equalize the number of oxygen molecules, and we do that by equalizing the volume by adding glass beads to the dry beads. Here's what it looks like:



Watch the video from 1:58 to 3:55

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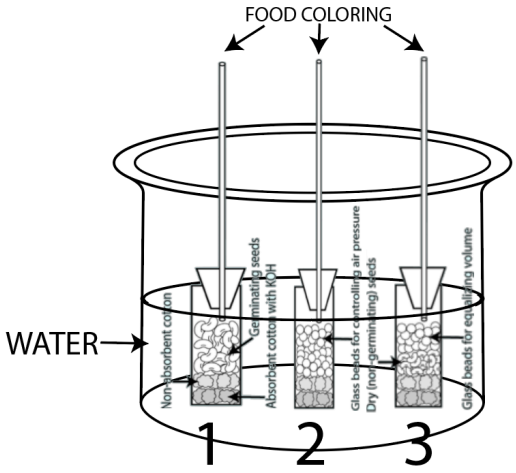
Cellular Respiration Lab: Sample Set Up

In this lab, we'd compare **four** things.

Germinating seeds have been given water. They're metabolically active.	1. Respiration rate of germinating seeds in at tap water temperature	2. Respiration rate of germinating seeds at ice water temperature
Dormant seeds are still dry. Their metabolic rate is very low.	3. Respiration rate of dormant seeds at tap water temperature	4. Respiration rate of dormant seeds at ice water temperature

…and we're trying to control for air pressure changes.



NOTE: All respirometers have absorbent cotton w/ KOH, and non absorbent cotton

BIG PICTURE:
Each team of four sets up 3 respirometers, either in ice water or in room temperature water.

SEED TYPE: LENTILS

SET UP

1. For respirometer 1: Get 50 germinating seeds (we'll decide based on which type of seed). Measure volume by displacement of water method. Record volume here: _____mL
2. For respirometer 2: Use beads to match the volume of germinating seeds. Set aside.
3. For respirometer 3: Use same number of *dried* (non-germinating) seeds. Measure their volume, using the displacement method. Add beads to match volume of germinating beans. Set aside.

Assemble respirometers

1. Place a small wad of **absorbent** cotton in the bottom of the chamber.
2. Using a 1ml plastic pipette, saturate the absorbent cotton using 1 mL of 10% KOH. Note: the KOH is a strong base. **Be careful.** Wear eye protection and rinse your hands off if any gets on your hands.
3. Add a small wad of non-absorbent cotton to protect your seeds.
4. Add *germinating seeds only* to respirometer 1.
5. Add *beads only* to respirometer 3.
6. Add *dry seeds and beads (to equalize volume)* to respirometer 2

Allow for *equilibration*: 1) Place respirometers in cups (one with ice, one without) Wait 3 minutes. Place a drop of food dye in the tip of your respirometer. Don't disturb. Allow 3 or four minutes before first reading. Things have equilibrated when the respirometer with beads (no organisms) has a relatively constant reading for a few minutes.

Keep temperature constant. During the course of the experiment, gently add ice to the ice cup as needed so that its temperature remains constant.

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Cellular Respiration Lab Analysis: SAMPLE DATA

DATA TASK I: Sample Data Using Lentils (**Everyone: complete this table.**

℃	Time (min)	Beads		Germinating Peas			Dry Peas and Beads		
		(a) Reading	(b) Difference)	(c) Reading	(d) Difference	(e) Corrected Difference	(f) Reading	(g) Difference	(h) Corrected Difference
25	Initial (0)	0.93	--------	0.91	--------	---------	0.92	---------	---------
	0 to 5	0.91		0.84			0.89		
	0 to 10	0.90		0.77			0.87		
	0 to 15	0.90		0.71			0.87		
	0 to 20	0.90		0.64			0.85		
10	Initial (0)	0.95	-------	0.92	--------	----------	0.91	---------	---------
	0-5	0.94		0.88			0.90		
	0-10	0.92		0.85			0.87		
	0-15	0.93		0.83			0.86		
	0-20	0.93		0.80			0.85		

DATA TASK II: Graph the corrected differences for all four treatments on a separate sheet of paper. Make a high quality photograph of your graph, and insert it below.

ANALYSIS QUESTIONS

Answer the questions below. Make your answers as complete as possible.

1. Explain the overall setup of the respirometer we used in this lab.
    a. How did it work?
    b. Why did we need to have KOH?
    c. Why did the food coloring move toward the the respirometer's chamber?
    d. Why was it necessary to correct the readings from the seeds with readings from the beads?

2. Think of the overall setup of this lab. What controls were used?

3. DESCRIBE and EXPLAIN the relationship between the amount of O_2 consumed and time.

4. What is *dormancy*? Explain what's happening in germinating seeds that's causing them to have a higher rate of respiration than dormant seeds.

5. Draw a sample graph of oxygen consumption for germinating seeds that starts at 5 degrees C and gradually heats up to 50 degrees C. Explain your prediction.

6. Before answering this next question, think for a minute about how the metabolisms of fish, amphibians, and reptiles is different from that of birds and mammals. The first three groups are commonly called "cold blooded." A better term is "ectothermic." Their body temperature matches the outside environment, and therefore fluctuates with environmental temperature. Birds and mammals are commonly called "warm blooded," but a better term would be "endothermic." Their heat comes from the inside, generated by cellular respiration.
   This has some big consequences. For one thing, the energy requirements of ectothermic and endothermic animals are quite different. For example, an 80 kg alligator can survive on about 100 food calories/day (or less), while an 80 kg person needs about 2000 food calories /day.
   Now, assume that we had a really big respirometer—big enough to hold a fence lizard (a reptile) and a field mouse (a mammal) of equal mass. If we compared their respiration rates at 10 ℃ (about 50 ℉), what results would you expect? Which animal would have the higher respiration rate? Justify your answer.

7. Using the same really big respirometer, imagine comparing the respiration rate of a mouse at 10 ℃ and a mouse at 25 ℃ (which is 77 ℉). Assume the mice have the same mass. Which animal would have the higher respiration rate? Justify your answer.

8. Your teacher has prepared four trays of germinating peas. In tray 1, the peas were just watered an hour ago (they're still dormant). In tray 2, they've been germinating for 24 hours. In tray 3, they've been germinating for 48 hours, and in tray four, for 72 hours. Using respirometers, design a controlled experiment to test the rates of cellular respiration in these different groups of peas. Explain how you would set up your expermiment, and propose a hypothesis of what the results might be. NOTE THAT THIS IS THE MOST IMPORTANT QUESTION IN THIS HANDOUT. *The goal is to set up a controlled comparison of these four groups of seeds. How would you do it?*