Cellular Respiration Lab by C. Kohn, Waterford WI

Name: Hour Date:

Date Assignment is due: Why late? Score: + ✓ -
 Day of Week Date If your project was late, describe why

# Lab Overview

In this lab, you will be using yeast to measure the rate of cellular respiration. Yeast is a single-celled eukaryotic organism that utilizes carbohydrates for ATP production in the same way that plant and animal cells utilize carbohydrates. Yeast will convert a carbohydrate into water and carbon dioxide during cellular respiration. The greater the carbon dioxide production, the greater the rates of cellular respiration. In groups of four, you will be determining the impact of temperature on cellular respiration by using a Waterford Fermenter to measure the CO2 production of the yeast. After completing the standard lab, you will be designing your own test of cellular respiration using an independent variable other than temperature and will measure its impact on cellular respiration.

# Needed Materials

Waterford Fermenter (7 for 24 students and instructor), tap water, a yeast packet (such as those sold for baking in grocery stores), an electrical outlet, titration materials (KOH, phenolphthalein, 37% hydrochloric acid, pipette or eyedropper, 1 molar strontium chloride (SrCl­2), and a graduated cylinder), a magnetic stirrer, safety equipment (gloves, goggles, and aprons). Note: you can also use a CO2 probe to determine carbon dioxide production in lieu of titration.

# Safety Warning

This lab involves dangerous laboratory chemicals – wear gloves, goggles, and aprons/lab coats at all times when conducting this lab. The chemicals used in this lab can produce dangerous fumes – work only in a well-ventilated area or a fume hood. If you spill a chemical on bare skin, flush immediately with lots of water while asking your instructor for help. Do not let chemical spills or broken glass go unnoticed – notify your instructor immediately if you have any spills or broken glassware.

# Itinerary

**Monday**: Review of material in groups; class discussion; summary of procedure

**Tuesday**: Protocol – effect of temperature on cellular respiration

**Wednesday**: Experimental design & set-up; class discussion

**Thursday**: Independent Experiments; Complete questions and lab reports

**Friday**: Oral Presentations; Peer Reviews

# Expectations

1. This is a lab that involves dangerous chemicals. Safety gear should be used whenever open chemical containers are present.
2. This lab requires you to follow instructions; all instructions are clearly written in this packet. Consult your directions first, then ask your instructor if you still have questions.
3. You will have to understand and think about the unit content to complete this activity. Use your brains!

Day 1: Review of Material Score /15

***Directions****: work with your group members to complete this sheet. Questions will be graded in class.*

1. What is the role of glucose (or other carbohydrates) in cellular respiration?

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2. What is the role of hydrogen in cellular respiration?
3. What is the role of ATP in cellular respiration?
4. What is the role of ADP in cellular respiration?
5. What is the role of ATP Synthase in cellular respiration?
6. What is glycolysis?
7. What is substrate-level phosphorylation?

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8. What is the TCA Cycle?
9. What is the role of NAD+/FAD+ in respiration?
10. What is the role of the Electron Transport System in respiration?

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11. What is oxidative phosphorylation?
12. What is the role of oxygen in respiration?
13. What are 3 possible ways to increase the amount of ATP produced by a cell? (1 pt each). **Describe in detail**!

1.­

2.­

3.­

Day 2 – Effect of Temperature on Respiration

# Overview

In this lab, you will be testing the effects of temperature on cellular respiration. Using yeast, a single-celled eukaryotic organism (i.e. yeast cells have organelles like plant and animal cells do), you will determine if temperature affects the rate of cellular respiration by measuring CO2 production in your heated fermenter and compare it to your instructor’s unheated fermenter. Using a Waterford Fermenter, you will increase the temperature of your yeast/sugar solution from room temperature to 37o C (body temperature). You will let your yeast digest the sugar in the solution for 15 minutes; during this time, you will see CO2 escaping through the airlock on your fermenter.

The airlock of the fermenter will have a KOH solution inside. As the escaping CO­2­ bubbles move through the KOH solution, it will change the pH of the solution and make it more acidic. After 15 minutes has passed, you will add SrCl2 to “freeze” the reaction of your KOH at its current pH. You will add a pH color indicator and then add a strong acid drop-by-drop to determine the actual pH change. The less acid you need, the more the KOH was acidified and the greater the CO­2 production.

CO2 will change the pH of the KOH solution, making it more acidic. The less acid needed to cause a color change, the more CO2 that was produced

# In a nutshell…

1. Yeast will eat sugar and produce CO­­2 which will bubble out through the airlock.
2. The airlock contains KOH (a strong base); CO2 will make the KOH more acidic
3. After 15 minutes passes, you will measure how much the pH of the KOH changes using a color indicator and acid

# Methods

1. Prior to starting, watch your instructor demonstrate the methods with their own fermenter and **cold** tap water. This fermenter will also serve as the control and will be left unheated.
2. For your group of 4, obtain a Waterford Fermenter and a packet of yeast.
3. Add 300 ml of warm (30-40o) tap water to your fermenter chamber; allow this water to sit in contact with your glass heater so that the temperatures of each can acclimate. Plug in your heater and heat to 380 C[[1]](#footnote-1)
4. Add the yeast and place the fermenter on a magnetic stirrer. With the magnetic stir bar in the chamber, turn on the stirrer and rapidly stir the solution for about 5 minutes so that the yeast become re-activated.
5. Answer the questions on the following page while your yeast is being re-activated.
6. After answering the questions, carefully add 25 ml of 1 M KOH solution[[2]](#footnote-2) to your fermenter’s airlock chamber (NOTE: you need gloves, goggles, and aprons and a well-ventilated area for this!).
7. Add 3 teaspoons of sugar to your yeast and water. Keep mixing the solution on the magnetic stirrer. Seal the fermenter securely so that it is airtight (air should only be able to escape through the airlock).
8. After 15 minutes has passed, add 2 ml 1 molar SrCl2[[3]](#footnote-3) to the KOH solution in the airlock (the clear chamber on top). This is necessary to “freeze” the reaction occurring between the KOH solution and the CO2
9. Add two drops of phenolphthalein. The solution should turn pink. Recap your phenolphthalein bottle.
10. Pour the KOH solution with SrCl2 and phenolphthalein into a small graduated cylinder.
11. Add 37% hydrochloric acid drop by drop until the pink color disappears. Gently swirl the KOH solution in between drops of acid. The less acid that is needed to turn the solution clear, the more CO2 that was produced.
12. Record this data and answer the remaining questions. Wash your equipment and allow it to dry.

# Pre-Experiment Questions (answer while your yeast is being re-activated)

1. Do yeast cells have mitochondria? How do you know?

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2. What will happen to the sugar when you add it to the yeast solution?

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3. What is your research question for this experiment? Write it below:

We are unsure if

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4. What is your hypothesis for this experiment? Write it below:

We predict that

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5. What is your rationale for your hypothesis? We think that that this will happen because

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6. What is your independent variable (the thing you purposely changed) for this experiment? Write it below:

Our independent variable is
7. What is your dependent variable (the thing you measure) for this experiment? Write it below:

Our dependent variable is
8. How will you measure your dependent variable? ATP production/CO2 production will be measured using titration; the less acid needed for a color change, the more CO2 / ATP that was produced
9. What is your control for this experiment?

**Results: Data & Observations (complete after your experiment is finished)**
10. What happened after you added sugar to your yeast/water solution?

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	1. Why do you think this happened?

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11. How many drops of acid did it take to change the color of your KOH solution?
12. How many drops of acid did it take to change the color of the control KOH solution in your instructor’s

fermenter?
13. Which fermenter required more acid to cause a color change, yours or the instructor’s?

	1. What does this indicate about temperature and respiration?

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	*Reminder that the fewer drops of acid needed for a color change (from pink to clear), the greater the CO2* *production.* ***More acid needed = less CO2* = *less respiration/less ATP produced; less acid needed = more CO2 /ATP***
14. Did anyone in your class have drastically different results than your own? Explain:

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15. Create a graph in the space below comparing the amount of acid you needed to use compared to the results of the control (your instructor’s fermenter).

# Conclusion (complete after your experiment is finished)

1. What was your original hypothesis? We predicted that

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2. What is your rationale for your hypothesis? We thought that that this would happen because …

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3. Was your hypothesis supported by your data?

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4. If you repeated your experiment again, do you think you would get the same results? Yes / No

Explain:

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5. Why do you think you saw the results that you did, and how do you think temperature affected cellular respiration? Answer by relating temperature to specific aspects of respiration (such as glycolysis, ATP synthase, etc.).
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Day 3 – Independent Experimental Design

# Overview

In this lab, you will be selecting an independent variable and measure its effect on cellular respiration. Suggestions will be provided for you; you can also choose a different variable

# Experimental Options

1. The effect of complex carbohydrates on rates of respiration
2. The effect of increasing the concentration of sugar on respiration
3. The effect of rapid agitation on respiration
4. The effect of aeration on respiration
5. Alternate option (per instructor’s approval)

# Methods

1. As a group, decide what experimental option most interests you. If you are selecting an option not listed above (i.e. the alternate option), speak with instructor and get their approval before continuing.
2. Complete the questions on the next page. Answer them as a group.
3. When you have completed these questions, speak with your instructor. If they are with another group, patiently wait until they are done.
	1. It would be a good idea to review your questions and plan if you are waiting to make sure you haven’t made any mistakes or left out any details.
4. Make sure that everyone is clear on what their responsibilities will be when you conduct the experiment and that everyone agrees to these plans.
5. Be prepared for a class discussion on what each group will be doing if time allows. While waiting, decide what each person’s speaking role will be during the presentation. Review the outline below and be prepared for questions!

# Oral Presentation

1. As a group, describe each of the following to the class:
	1. Independent variable – what you intend to change
	2. Dependent variable – what you intend to measure
	3. Question – what are you trying to determine?
	4. Hypothesis – what do you predict will occur?
	5. Rationale – why do you think your hypothesis will be correct?
2. Allow time for questions – your instructor or other students may ask the following:
	1. Why do you think your experiment will be a good test of your hypothesis?
	2. Why do you think your expected outcome will occur?
	3. What is your control?
	4. What factors might throw off your experiment and change its results?
	5. Could the exact opposite of your hypothesis occur? What might make that happen?
	6. Do expect you would get the same results each time? What might make your results more variable?
	7. Are there any details that you did not consider that might affect how others would conduct this experiment?

# Pre-Experiment Questions (answer BEFORE asking for instructor approval)

1. Briefly describe what you intend to do in this experiment:

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1. What is your research question for this experiment? Write it below:

We are unsure if

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2. What is your hypothesis for this experiment? Write it below:

We predict that

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3. What is your rationale for your hypothesis? We think that that this will happen because

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4. What is your independent variable (the thing you purposely changed) for this experiment? Write it below:

Our independent variable is
5. What is your dependent variable (the thing you measure) for this experiment? Write it below:

Our dependent variable is
6. How will you measure your dependent variable?

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7. What is your control for this experiment? Our own earlier results from Day 2
8. What materials do you need for this experiment? List ALL supplies and materials needed below:
*Hint: you may want to look at the materials used earlier in this packet for guidance*
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9. What are your methods for this experiment? Write them below in a “cookbook” step-by-step style:
*Hint: you may want to look at the methods used earlier in this packet for guidance*\_

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10. What will be the role of each person during this experiment?

Person 1: Responsibilities:

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Person 2: Responsibilities:

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Person 3: Responsibilities:

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Person 4: Responsibilities:

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**Instructor’s Signature of Approval: Date:**

# Results: Data & Observations (complete after your experiment is finished)

1. What were the results of your experiment?

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	1. Why do you think this happened?

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2. How many drops of acid did it take to change the color of your KOH solution?
3. How many drops of acid did it take to change the color of your control fermenter?
4. Which fermenter produced more CO2, yours or the control?

	1. What does this indicate about your tested variable & respiration?

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	*Reminder that the fewer drops of acid needed for a color change (from pink to clear), the greater the CO2* *production. More acid needed = less CO2* = *less respiration*
5. Create a graph in the space below comparing your results to the results of the control:
6. Write a caption for this graph (*a caption should describe what the graph shows and what that means)*:

In this graph you can see that

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# Conclusion (complete after your experiment is finished)

1. What was your original hypothesis? We predicted that

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2. What is your rationale for your hypothesis? We thought that that this would happen because …

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3. Was your hypothesis supported by your data?

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4. If you repeated your experiment again, do you think you would get the same results? Yes / No

Explain:

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5. Why do you think you saw the results that you did? Answer by relating your tested variable to specific aspects of respiration (such as glycolysis, ATP synthase, etc.).

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Peer Review – Agricultural Sciences

**Name: Date: Hour:**

**Directions**: Please evaluate your group as well as yourself on the basis of contributions and effort on a scale of 1 to 5. A group member who makes an outstanding contribution and did their best would receive a score of 5. A group member who did very little might score around a 3, and a group member who did little or nothing might get a one or a two. Be sure to provide a reason for your score – why did you assign the score that you did? *(5’s need no reason)*

**1. Group Member’s Name: Score: 1 2 3 4 5**

Reason:

**2. Group Member’s Name: Score: 1 2 3 4 5**

Reason:

**3. Group Member’s Name: Score: 1 2 3 4 5**

Reason:

**4. Your Name: Score: 1 2 3 4 5**

Reason:

**Additional comments or concerns:**

 **Changes you would recommend for this activity:**

1. The heater is only on or off; if the orange light is on, it is heating. Adjust the knob on top so that the light turns off at 38o [↑](#footnote-ref-1)
2. Instructors: To make 1 M KOH, dissolve 1.1 g KOH in 100 ml of water in an Erlenmeyer flask or beaker. [↑](#footnote-ref-2)
3. 1.6 g SrCl2 per 10 ml of water [↑](#footnote-ref-3)