

# Cellular respiration



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## LABORATORY REPORT FOR BIO411 Experiment 5 : Cellular Respiration

Experiment 6 : Photosynthesis Title: Cellular Respiration Objective: To observe and determine cellular respiration in yeast/onion cells. \* Measure respiration rate using different substrates. \* Measure respiration rate at different temperature.

Introduction: In this laboratory experiment, we are given 3 task. The first one is respiration in yeast. Second is respiratory indicator and the third one is observing mitochondria in yeast/onion cell. The first experiment is about investigation of sugars yeast. We will determine which sugars yeast can be use for cell respiration. When the yeast undergoes anaerobic/aerobic respiration, they will give out carbon dioxide. After that it will reacts with water to forms a weak carbonic acid solution. We will use BTB to monitor this reaction.

Further explanation will be discussed at the discussion. The second experiment is respiratory indicator. Same concept with the first experiment. We will measure the respiration rate using different substrates based on the table provided.

The third experiment is observing mitochondria in yeast/onion cells. We will be using the staining technique in order to obtain the result of the experiment. \* Task 1: Respiration in Yeast Materials: Beakers \* Pipettes \* Cuvettes \* 20% Glucose \* 20% Lactose \* 20% Sucrose \* 20% Maltose \* Distilled water \* Brom Thymol Blue (BTB) \* Spectrophotometer \* Measuring cylinders. Procedures: 1. The spectrometer is set to 565nm. Distilled water is used to set the reading to the pure level.

2. 8mL of 20% glucose is placed in the beaker using a measuring cylinder. 3. 1ml of Brom Thymol Blue (BTB) is added into a respective tubes using a pipette. 4. 4ml from the mixture of Glucose and BTB taken and placed in test tube then added with 0. 1ml of yeast extract. 5.

The mixture then transferred into cuvette and placed into spectrophotometer. 6. The absorbance measured for 5 minutes. The reading taken within 30 seconds.

7. Steps 1-6 is repeated using 20% Lactose, 20% Sucrose, and 20% Maltose.

8. Reading obtained is recorded and compared with the others. Results: Time (s)| 20% Glucose| 20% Sucrose| 20% Maltose| 20% Lactose| 30| 1. 053| 1.

162| 1. 323| 1. 130| 60| 1. 051| 1. 161| 1. 310| 1. 123| 90| 1. 049| 1.

159| 1. 304| 1. 119| 120| 1. 048| 1. 159| 1. 297| 1.

116| 150| 1. 045| 1. 158| 1. 295| 1. 114| 180| 1. 044| 1.

155| 1. 292| 1. 111| 210| 1. 041| 1. 155| 1. 289| 1. 109| 240| 1.

040| 1. 154| 1. 287| 1. 107| 270| 1. 037| 1. 154| 1. 287| 1. 105| 300| 1.

035| 1. 154| 1. 284| 1. 103| Task 2: Respiratory Indicator Materials: \* Test tubes \* Yeast extract \* 20% Glucose \* Water bath \* Parafilm \* Distilled water \* Tap water \* Methylene blue Procedures: 1. 4 test tubes is obtained. 2. Each tube filled as table shown below, Tube 1 (room temperature)| Tube 2 (100? c)| Tube 3| Tube 4| 5ml yeast+1ml glucose+2ml methylene blue+2ml distilled water| 5ml yeast+1ml glucose+2ml methylene blue+2ml distilled water| 5ml water+1ml glucose+2ml methylene blue+2ml distilled water|

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5ml yeast+1ml water+1ml methylene blue +2ml distilled water| 3. For tube 2, yeast is added and glucose is immersed the tube in water bath(100%) for 5 minutes.

The, methylene blue is added. 4. Parafilm is used to cover all the tubes. 5. Initial time and color is recorded. 6.

Time taken for the decolorisation to occur is recorded. Result: | Tube 1| Tube 2| Tube 3| Tube 4| Observation| Dark blue(no change)| Dark blue(no change)| Dark blue(no change)| Dark blue(no change)| \* Task 3: Observing

Mitochondrion of Onion Cells Materials: \* Glass slides \* Sucrose solution \* Methylene blue \* Toothpick \* Onion cell \* Cover slip \* Microscope

Procedures: 1. Clean slide is obtained and a drop of sucrose solution is placed on the center.

Two drops of methylene blue added and mixed well by using toothpick. 2. Yeast is placed on the mixture of sucrose and methylene blue and covered by the cover slip. 3.

The slide immediately viewed under microscope. Results: 35 minutes is taken for yeast cells to decolorized, where blue stain turns clear. Discussion: In our experiment, in order to achieved our objective which are to observe and determine cellular respiration in yeast/onion cells, we have to measured respiration rate using different substrates and we also have to measure respiration rates at different temperatures in Task 1, Task 2, Task 3. In Task 1: Respiration in yeast, the result we obtained shows that the 20% Glucose give the lowest value of absorbance compared to the others and 20% Maltose gave the highest value of absorbance.

The order of absorbance is 20% Glucose <20% Lactose <20% Sucrose <20% Maltose. This shows that Maltose has the highest rate of sugar which has the effective for yeast respiration. The wavelength used is same, but the reading was taken in every 30 seconds in 5 minutes period of time. As known, cell released energy from the food molecules by process of respiration. Cell also can dissolve in water to form a weak acid. As result, pH indicator such as BTB can be used to indicate the presence of carbon dioxide. “ At low temperatures (0-10 C) yeast will not grow, but not die either. At temperatures 10-37 C yeast will grow and multiply, faster at higher temperatures with an optimal growth at 30 or 37 C (that depends on the species).

At higher temperature the cells become stressed, meaning that their content becomes damaged and which can be repaired to some degree. At high temperatures (> 50 C) the cells die. The bacteria can survive freezing under certain conditions. When baking bread all yeast dies during the process. ” - Dr. Trudy Wassenaar  
In Task 2: Respiration indicator, the result we obtain is no change at all the tubes prepared according to the table provided.

Tube 1 supposed to have change in their colour because it was occur in room temperature, consist of yeast, have glucose and distilled water. No change for Tube 2, 3 and 4. Tube 2, it was occurred in 100 Celsius. Thus, no change occurred. For Tube 3, there are no change in colour because it does not consist of yeast, and for Tube 4, no change also because absent of glucose. For tube 2, no change occur because yeast can survive only in certain condition, as the researcher explained below:-  
In Task 3: Observing mitochondria in yeast/onion cells, we used yeast as our specimen. It actually  
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takes 20-35 minutes to decolorize the blue stain. Unfortunately, we does not managed to observe the bluish oblong bodies in cytoplasm of cells.

This maybe because the magnification we used is still cannot managed to give a clear image of the mitochondria. Conclusion: The objective is successfully achieved which is to observe and determine cellular respiration in yeast using different substrates and temperatures. Reference: 1) [http://www.newton.dep.anl.gov/askasci/bio99/bio99693.](http://www.newton.dep.anl.gov/askasci/bio99/bio99693.htm)

htm \* LAB 6 Title: Photosynthesis Introduction: In this experiments, we will mostly will discuss about process of photosynthesis which is process where the plants will convert carbon dioxide into organic compound. We have given 3 task in this experiments. There are:- Task 1, we have to extract chlorophyll pigments using paper chromatography, the separation technique. When the absorbent paper has its end puts into a solvent, the solvent will creep up the paper and soak it.

Solute in solvents also tend to be carried upwards with the solvent. Various types of chlorophylls and carotenoids of plant photosystems are all membrane bound and only soluble in rather non polar solvents. These pigments have been removed from the spinach by extraction into acetone. The pigments will be separated by chromatography by using a solvent. Task 2, we have to measure wavelength of light absorbed by plant chlorophyll. Spectrometre is used to determine the absorbtion spectrum for the available chlorophyll extract of spinach pigments.

Absorbance from the wavelengths of 400nm to 720nm every 10nm.

Methanol used as a blank. Task 3, we have to quantifying the amount of

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oxygen being released from the photosynthesizing solution or the more accurate is we have to determine the rate of photosynthesis. Task 1:

Photosynthesis Pigments Objectives: To extract chlorophyll pigments using paper chromatography. Materials: \* Fresh spinach \* Methanol \*

Chromatography paper \* Conical flask \* Rubber bugs \* Haematocrit needles \* Pencil \* Ruler \* Spectrophotometer Procedures: 1.

A strip of absorbent paper positioned that it suspended about 5mm from the bottom of the available flask. 2. The paper is removed and acetone is poured into the bottom of the flask so it will be touching the bottom of the paper. 3. A strip of absorbent paper is streaked with the spinach extract using the haematocrit.

4. Dry it in a moments. A second streak is applied over the first one. It repeated for minimum 15 times. Then dry it. 5.

The paper is hooked on the stopper. The paper inserted into the flask so that its bottom just immersed into the solvent. 6. Let it until the solvent reached pin.

7. The diagram of the pigmentation on chromatography paper, labelled.

Result:  $R_f = x / y$  Where, x is 9.9 cm and y is 15.2 cm. Thus,  $9.9 / 15.2 = 0.611$  cm ( Not accurate- Experiment failed) \* Task 2: Absorption

Spectrum Objective: To measure the wavelength of light absorbed by plant

chlorophyll. Materials: \* Beaker Acetone Procedures: A spectrophotometer

used to determine the absorption spectrum for the available chlorophyll extract of spinach pigment. The absorbance measured from wavelengths of

400nm to 720nm in every 10nm. Methanol used as a blank. Result:

Wavelength ( ? ) | Absorbance | Wavelength( ? ) | Absorbance | 400 | 0.251 | 570 | 0.

224 | 410 | 0.262 | 580 | 0.221 | 420 | 0.255 | 590 | 0.

226 | 430 | 0.260 | 600 | 0.235 | 440 | 0.271 | 610 | 0.220 | 450 | 0.

260 | 620 | 0.223 | 460 | 0.259 | 630 | 0.221 | 470 | 0.259 | 640 | 0.220 | 480 | 0.251 | 650 | 0.216 | 490 | 0.

251 | 660 | 0.214 | 500 | 0.248 | 670 | 0.

213 | 510 | 0.27 | 680 | 0.250 | 520 | 0.229 | 690 | 0.220 | 530 | 0.

242 | 700 | 0.266 | 540 | 0.274 | 710 | 0.237 | 550 | 0.233 | 720 | 0.198 | 560 | 0.230 | | | Table 1.

1 | \* Task 3: Measuring Rate of Photosynthesis Objective: To determine the rate of photosynthesis. Materials: \* Elodea plant \* 10mL measuring cylinder \* Test tubes \* Beaker \* 0.5 sodium hydrogen bicarbonate buffer Procedures: 1. Fresh sprigs of Elodea plant is inserted into wide test tube. The tube fully filled with buffer solution. The pH of buffer taken. 2.

The tube was inverted into beaker which is half filled with the same buffer solution. 3. There is must be no air space inside the tube. 4. The beaker placed under a table lamp for one hour.



5. The tube gently tapped, to trapped air bubbles released from the leaves.

6. The bubbles released counted. 7. The pH obtained recorded. Results:

Reading| Initial| Final| pH| 8.

02| 9. 31| Volume| 25 ml| 24 ml| Total bubble formed= 4 Discussion: In our experiment, we have to discuss about the photosynthesis. What is photosynthesis? Photosynthesis is essential process for any living plants to survive. Raw materials which are carbon dioxide, water, oxygen and glucose as the products of the reaction. Water molecules will be broken down and oxygen will be released. Glucose molecule contains the energy from the sunlight converted to a new form, chemical energy. The overall chemical equation for these reactions is shown below:  $6 \text{CO}_2 + 6 \text{H}_2\text{O} + \text{energy} \rightarrow \text{C}_6\text{H}_{12}\text{O}_6 + 6 \text{O}_2$  The energy that plants trap is essential, both for their own growth and for other organisms that rely on plants for food.

Pigments that involved in this process is chlorophyll. So as in Task 1, we had extracted the chlorophyll pigment in spinach using the paper chromatography. Paper chromatography is a useful technique for separating and identifying pigment and other molecules from cell extracts that contain a complex mixture of molecules).

We may have the result stated as below but to be truth is our experiment does not have an accurate result because of the spinach with the acetone is too dilute and it is difficult to obtain an accurate result. It was suppose to separate the pigments in paper chromatography but, we only can observe a single solute carried upwards. In Task 2, which is we measure the wavelength of light absorbed by plant chlorophyll starting from 400nm till

720nm. Reading is taken every 10nm. Based on the Table 1. 1, we can see the absorbance of chlorophyll are in a range of 0. 198 and 0. 274.

There are not much different of absorbance in wavelength of 400nm until 720nm, but it the absorbance seems like decreasing where 0. 251 at 400nm, and 0. 198 at 720nm. In Task 3, we have to determine the rate of photosynthesis. In this experiments, we used Elodea plant.

(Elodea is an autotroph that will photosynthesize under the appropriate conditions. )The initial pH value of buffer is 8. 02, and the initial volume is 25 ml. After 1 hour, the pH value increased to 9.

31, but decrease of volume to 24 ml. The pH increased because of higher amount of carbon entered the intermediate pathways. Total bubble produced was 4. The bubble produced means the total oxygen produce by the plant in photosynthesis process.

Conclusion: We have achieved the all the objective of experiments which is to study Photosynthesis in 2 Task, whereby we need to extract chlorophyll pigments using paper chromatography, measure the wavelength of light absorbed by plant chlorophyll and to measure rate of photosynthesis.

Reference: 1) [http://www. ucmp. berkeley. edu/glossary/gloss3/pigments. html](http://www.ucmp.berkeley.edu/glossary/gloss3/pigments.html) 2) [http://en. wikipedia.](http://en.wikipedia.org/wiki/Photosynthetic_pigment)

[org/wiki/Photosynthetic\\_pigment](http://en.wikipedia.org/wiki/Photosynthetic_pigment)