



Name: _____ Date: _____ Per: _____

Lab: The Effects of Environment on Gene Expression

Background Information:

Every living organism contains genetic material--such as DNA. Our DNA is expressed in the processes of transcription and translation, during which proteins are created that determine many of our traits. Sometimes, factors such as environmental changes, can play a role in the way our genes are expressed.

One environmental factor that can influence gene expression is sunlight. Any organism that derives its energy from light and can use carbon dioxide as its principal source of carbon is photoautotrophic. Examples of photoautotrophs include plants (ex: fern) as well as some bacteria (ex: cyanobacteria) and some protists (ex: algae).

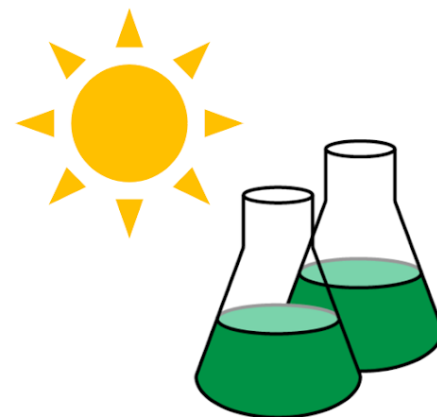
Any organism that can use only organic materials as a source of energy is heterotrophic. Examples of heterotrophs include animals, fungi, and most bacteria and protists. Sometimes an organism can be both autotrophic and heterotrophic depending on their environmental conditions. If sunlight is not readily available, then an organism may be able to create energy in a heterotrophic manner. This change in environment, light availability, can influence the way that an organism's genes are expressed into traits.

Pre-Lab Questions:

1. What is one way (other than sunlight availability) that the environment affects living organisms? Give a specific example of how the environment influences the way a living organism expresses its traits.
2. When was a time that the environment changed the way you feel/ act/ look? Did it result in a permanent change or a temporary one? Do you think your DNA was altered as a result of the environmental change--why or why not?

Materials:

- Growth media (Ex: BOLD media)
- Green algae (*Chlamydomonas reinhardtii*)
- Erlenmeyer flasks or culture flasks (250-500mL)*
- 91% isopropyl alcohol (other options: acetone, ethanol)
- Silica chromatography paper
- Microfuge tubes
- Centrifuge
- Micropipette and tips
- compound light microscope, slides & cover slips
- Hemocytometer



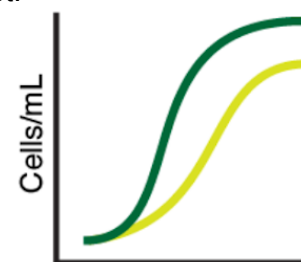


- **OPTIONAL:** Spectrophotometer or Vernier SpectroVis

***NOTE:** If using Erlenmeyer flasks, you can use aluminum foil as a cover OR make your own stoppers that allow for gas exchange using cotton balls, cheesecloth and tape.

Procedure:

1. Obtain 2 Erlenmeyer or culture flasks and label them "Light" and "No Light".
2. Add growth media (ex= BOLD media) to each flask to fill it about $\frac{1}{3}$ full.
3. Inoculate your flasks with about 10% of its total volume worth of algae from your teacher's stock culture. (ex: if you have 100mL of growth media, inoculate with 10mL algae). Immediately cover your flasks with a cap (if provided), stopper, or aluminum foil.
4. Perform an initial cell count of your algae that you just inoculated. Make sure
5. Pellet down 1mL of suspended cells in TP media using a microfuge tube and centrifuge.
6. Decant off the supernatant, keeping the pellet.
7. Add 1-2 mL 91% Isopropyl alcohol and resuspend the cells within the pellet.
8. Using a spectrophotometer or Vernier SpectroVis to measure the absorbance of the sample at 680 & 720nm, blanking with isopropyl alcohol, and record values.
9. Leave 1 flask labeled "Light" with access to light and the other flask labeled "No light" inside of a dark cabinet. Make sure to label with group names and to put flasks exactly back where they were after gathering data points.
10. Repeat steps 2-6 for a week, collecting data for a sample of cells left with access to light and for those without light.
11. Calculate chlorophyll per cell by graphing OD_{680} vs. time including both sets of data (light & dark) on the same graph. Draw 2 best fit lines for each set of data, making a key for each set up. Attach your graph separately.
12. **If time:** Use a micropipette tip or capillary tube and make dots of pigment from your samples in light vs. dark on a silica thin layer chromatography plate. You can use the 91% isopropyl alcohol as an eluent. Wait. Leave the paper to run for at least 15 minutes, checking it periodically for pigment separation. Draw your results and discuss if you see any differences in chlorophyll.



Hypothesis:

Formulate a scientific hypothesis below about how the two flasks you set up, with and without access to light, may vary throughout the experiment. After your hypothesis, write a rationale or a "because" statement that explains why you think that may occur.

Define Variables:

Controlled variables=

Independent variable=

Dependent variable=



Data Collection: (modify as needed)

Green Algae Chlorophyll Production in Light:

Date	Cell Count (cells/mL)	OD ₆₈₀	OD ₇₂₀

Green Algae Chlorophyll Production in Dark:

Date	Cell Count (cells/mL)	OD ₆₈₀	OD ₇₂₀

Conclusion Post-Lab Questions:

1. Based on the data from your experiment, how does a change in environment (light in this case) affect gene expression in *Chlamydomonas reinhardtii*? Support your answer using your data.
2. When grown without some nutrients, like nitrogen for example, green algae like ours used in this lab, have been shown to produce oils that can be extracted and used for biofuel. How is this scenario similar to the lab you just performed?
3. Was your hypothesis supported? Why or why not? Were there any factors other than the independent variable that may have inadvertently influenced your results?



EXTENSION: 1) Perform the experiment again, but with a different independent variable other than light--design your own experiment testing 1 change in the environment of the algae. Make sure you can measure both qualitatively and quantitatively how gene expression may alter in response to your changed variable. 2) Perform the same experiment, but instead of extracting chlorophyll, grow the algae in flasks with varying distances from a constant light source--measuring photons if possible, and with a control group in the dark. You could test to see if there is an optimal distance from a light source.