**Environmental Influence on Gene Networks - Advanced Prep for the Module**

Entry Behaviors

* Understand how genes and proteins are related.
* Know specifics of gene transcription and translation.
* Know the molecule ATP, be familiar with its use in a cell and its components.

Required Lab Equipment:

* Spectrophotometer with cuvettes (if you want to look at quantified cell growth).
  + There are ways you can borrow these if you plan ahead. For example, Washington State University has a free equipment loan program (they do have spectrophotometers). Here is the link - [http://www.sci.wsu.edu/bio/hhp/index.html](http://livepage.apple.com/). Or you can borrow them from the ISB through SEE. Contact Claudia Ludwig for more information: [cludwig@systemsbiology.org](mailto:cludwig@systemsbiology.org).
* Microcentrifuge
* Full Spectrum Flood Light (must be full spectrum)
  + Home Depot sells these – specifications: PAR 38 with 90-100 watt and 1370-1500 lumens.

You many need multiple of these lights. The better your light placement, the better the bR production.

Suggested Equipment:

* Computers: If you don’t have access to student computers, you will need to print references for Lesson 1. Additionally for Lesson 4’s Purple Membrane (bR) simulation, the teacher will need to use a demo computer.
* Agitating Incubator to use Falcon tubes OR stir stations to accommodate 4, 50 mL flasks per group.
  + If your incubator doesn’t agitate – you can use small flasks with stir bars.
  + If you have an incubator but no stir stations, periodically agitate the containers manually and increase the incubation time 24-48 hours. Use 1.5mL (full microcentrifuge tube) sample when you spin down the cells.
  + If you do not have an incubator – you can use stir stations AND the light for heat.
  + If all you have are lights, agitate the sample containers a couple of times a day and you will still get results. Plan on letting the samples grow 1 or 2 more days. If you can’t see a clear difference in light vs. dark after spinning down the cells, try using 500μL instead of 1.5mL when you spin down the cells. Agitation increases the growth rate so this can actually cause the samples to appear a darker color than they actually are.

Overall Module and Lesson Timeline:

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| Lesson # | # of 50 min Class Periods | Timing Notes |
| 1 | 1 | This lesson could be made a homework assignment if the teacher only has time to give pre-assessment and introduce module. |
| 2 | 3 | Lab needs to have incubation time of 72 hrs. Easiest way to get this is if the students set-up their experiments and start their incubations on a Friday. This means ~72 hrs will have passed before they see you Monday. |
| 3 | 2 | The beginning of this lesson is the best point at which to put any review required, based on pre-assessment. |
| 4 | 3 | If you only have 1 centrifuge and/or one Spec, you will need to spread out your students’ result gathering (so you don’t have 7 odd groups waiting around for 1 group to get done). This is probably best accommodated either scheduling students to come after class OR during the first part of Lesson 3.  NOTE – if you do plan to have your students take their results at different times, you must stop everyone’s experiment at one time (to allow groups to combine/compare data). This can be done by just putting your sample on a lab bench (at room temp – OK as long as samples don’t sit longer than 24hrs). If the students will take longer than a 24hr window to get data, put sample in the refrigerator.  Depending on teacher’s plan for student presentations, more time may be needed. |

This timeline is the quickest timeline, teacher may require more time in certain areas.

**Recipe for making your own complete media (CM) for optimal *Halobacterium* growth**

For 1 liter of 4.3M stock solution, mass the following and place in a 1 liter container. Then add deionized (or distilled) water to make 1 liter. This is almost saturated with salt so will take a while to dissolve. Also, the peptone will clump and stick so will need to be stirred. Using a stir station will help.

|  |  |
| --- | --- |
| **Chemical** | **Amount** |
| salt (NaCl) | 250g |
| Magnesium sulfate heptahydrate (MgSO4.7H2O) | 20g |
| Potassium chloride (KCl) | 2g |
| Sodium citrate dihydrate (Na3C6H5O7·2H2O) | 3g |
| Peptone oxoid (neutralized bacterial amino acids) | 10g |

Reminder: when making solutions, the ingredients take up space so the solvent (water) is always added last. Final total volume should be measured to ensure 1.0L after all ingredients are in solution.

\*See Teacher Lab Hints in lesson 2 for more detail.