Evolution and Biodiversity Laboratory
Activity: Project Development and Protocol Instructions

When all of your team members have finished identifying organisms, it's time to convene and discuss your literature searches. These should give you ideas about the research project your team will now design and undertake for the next four lab sessions.

I. Developing Your Team Research Project

Review the Biodiversity Background Information Chapter of the online lab manual (linked to this week's session, too) for details of what is expected of you for this project.

By the end of today's lab session, your team should have a fully developed research project ready to begin next week.

Use the Research Protocol Template linked to the online syllabus to outline your project. Your team must submit the completed form to your lab instructor before you leave class today.

II. Instructions for Sampling Protocols

Your team will receive a sampling kit (Figure 1) and other collecting tools:

- **twelve plastic cups with lids**
- **plastic disposable pipets for each of your two localities**
- **disposable plastic straws (for sampling; Figure 2)**
- **pH strips (optional)**
- **one meter colored transect lines (you will place indelible marks at 20cm intervals)**
  - Use one cord each of your two habitats, but make sure they are **identical**.
  - Use an **indelible marker to place marks at 20 cm** intervals along the transect cords.

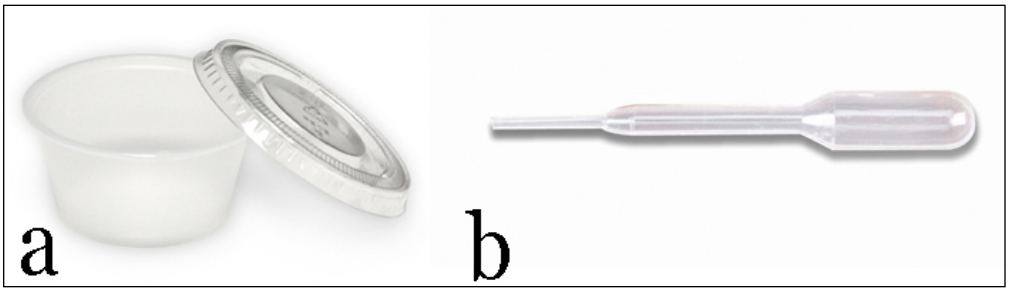

Figure 1. Plastic cup (a) and plastic pipet (b) are handy field collection tools.

When collecting samples, wear protective, disposable plastic gloves, and avoid direct contact with water. Unfortunately, local aquatic habitats are not always clean. Use due caution.

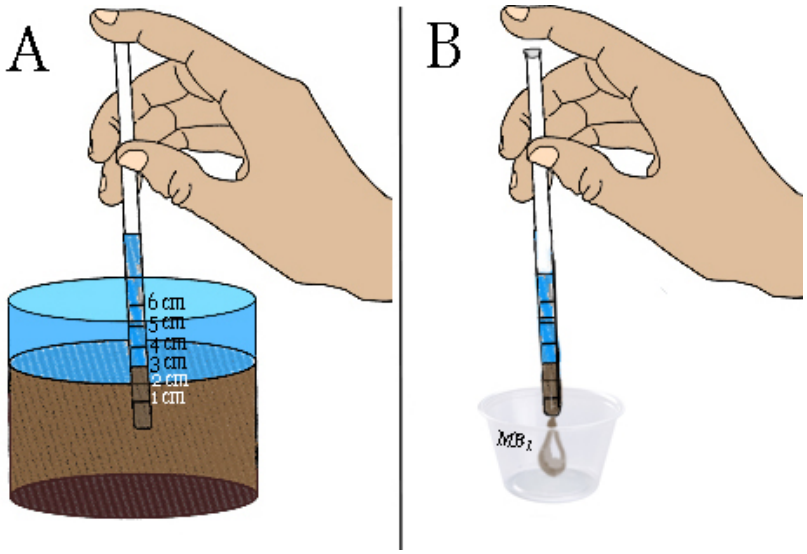

Figure 2. You can mark your collection straw with gradations and use it to sample sediment or water column (A) at specific depths. Dip the straw to the desired depth, and place your finger over the top of the straw to pull it up. To release the sample into your labeled sample cup, lift your finger from the top of the straw (B).

Before you come to lab next week, you must collect samples from BOTH of the selected habitats you wish to compare.

***Collect your samples as close to lab time as possible*, so the samples will be fresh and the organisms alive, kicking, and truly representative of what is living at that locality**

A. Preparing Your Sampling Kit

Next week, as close to lab session time as possible, you must visit each of your two ecosystems to collect samples.

1. You will use **six sample cups this week** (three for each habitat) and **six next week**.
2. Each week, **three cups will be used to collect samples at each site.**
3. **LABEL PIPETS AND STRAWS AND DO NOT USE THE SAME ONES TO COLLECT SAMPLES FROM DIFFERENT HABITATS! CROSS CONTAMINATION WILL AFFECT YOUR RESULTS.**
4. Label each of your 12 sample cups appropriately with
   - **a. Locality**
   - **b. Date**
   - **c. Transect #**
   - **d. GPS coordinates**

B. Using Your Sampling Kit

For this project, all teams will use the same sampling procedure and collect the same volumes of water, sediment, or soil (the nature of your project will determine which of these you collect.) along their transects. Here's how.

When you arrive at your first habitat, select **three collection sites**, making sure their physical features are relatively uniform and representative of the site. They should not be too distant from each other, but far enough apart to be representative of the habitat type.

1. Select a specific area in your first collection site.
2. Lay your transect cord along the area to be sampled, and pull it straight
3. As described above, choose a set distance from the line with a ruler.
4. **At that distance, at five different 20cm marks along your transect, collect**
   - **~6mL (water or sediment) if your habitat is aquatic**
   - **~1cc (soil or other solid) if your habitat is aquatic**
5. ***Place all five 6mL (or 1cc) samples from one transect into the same sample cup*.**
6. *Total*: 30mL (aquatic) or 5cc (terrestrial) per sample cup
7. **This is your first sample/replicate.** (The five contributions are subreplicates.)
8. Label the cup and its lid appropriately (locality, transect #, date, time, etc.)

You will perform the above procedure three times in each of your habitats.
Select a new location for each of the three transects you sample in each habitat.

Water/Soil Chemistry Analysis

If you want to be sure to have enough extra water/sediment/soil for performing chemical analysis, then collect ~ 30mL additional water/sediment (aquatic habitat) or ~10cc soil (terrestrial habitat) from any point along each transect and place in a separate lidded container. *Label the extra appropriately, so you know which transect it came from*.

In the lab, you will have resources available to test such abiotic parameters as

- pH
- nitrite
- nitrate
- ammonia
- phosphorus
- etc.
- (If you wish to test other parameters, check a local aquarium supply store or even Home Depot for options.)

At the site, record relevant environmental variables, such as

- air temperature
- water temperature
- weather conditions
- pH (strip kits available with sampling kits)
- light condition/quality
- etc.

Note whether these are different from one week (or one locality) to the next.

Remember that these abiotic variables will NOT necessarily allow you to pinpoint a cause for any differences in biodiversity you see between your habitats. But it may give you a starting point to generate ideas for further research.

Your team will repeat this procedure three times in each habitat.
You will do this for two weeks in a row on your lab session day.
When you have completed both weeks' data collection, you will have collected and analyzed a total TWELVE replicates, SIX from each locality.

At the end of WEEK ONE, you should have

- **Three replicates (sample cups) from Habitat #1**
- **Three replicates (sample cups) from Habitat #2**

At the end of WEEK TWO, your collecting saga will have yielded a TOTAL of

- **Six replicates (sample cups) from Habitat #1**
- **Six replicates (sample cups) from Habitat #2**

HINT FOR SUCCESS:
Keep sample cups capped only when traveling, to avoid spilling them.
Whenever possible, keep the lids ajar to allow oxygen to mix with the water.

Dead organisms are no fun to watch.

C. Recording Field Data

Before going to your collection localities, your team should devise an appropriate table for recording environmental data, such as that shown in Table 1.

Table 1. Example of a data collection table. In this example, the first habitat was sampled with a transect line traversing the intertidal sand sediment at Biscayne Nature Preserve (BN). The second habitat was sampled along a transect line traversing the sand sediment at the Matheson Hammock marine swimming pond (MH). Three replicates/samples were collected at each habitat in each week for a total of six samples/habitat

Sample #	Date	GPS	Air temp (°C)	H_2O temp (°C)	Time	Notes
BN 1	8 Feb 2019					
BN 2	8 Feb 2019					
BN 3	8 Feb 2019					
BN 4	15 Feb 2019					
BN 5	15 Feb 2019					
BN 6	15 Feb 2019					
MH 1	8 Feb 2019					
MH 2	8 Feb 2019					
MH 3	8 Feb 2019					
MH 4	15 Feb 2019					
MH 5	15 Feb 2019					
MH 6	15 Feb 2019					

The above is only a sample. You will probably wish to include more information in yours.

HINT FOR SUCCESS:
Whenever possible, photograph your site and your team in action.
Photos will enhance your final presentation.

D. Next Week: Bringing Your Samples to Lab

Your team should plan to collect samples no more than a few hours before lab.

Some organisms are more tolerant of disturbance than others.
The composition of organisms in your sample can change with time, as more sensitive organisms die. This will obviously affect your data! Prevent it as best you can.

Once a sample is collected, keep its temperature and lighting conditions as close to those at the sampling site as possible until you bring it to lab for analysis.

You will have two weeks for data analysis. If there are changes in weather or other environmental conditions between dates, be sure to record those in your field notebook, in case you wish to refer to them to help explain your results.

The wise ecologist records *anything* that can affect the nature of collected data.