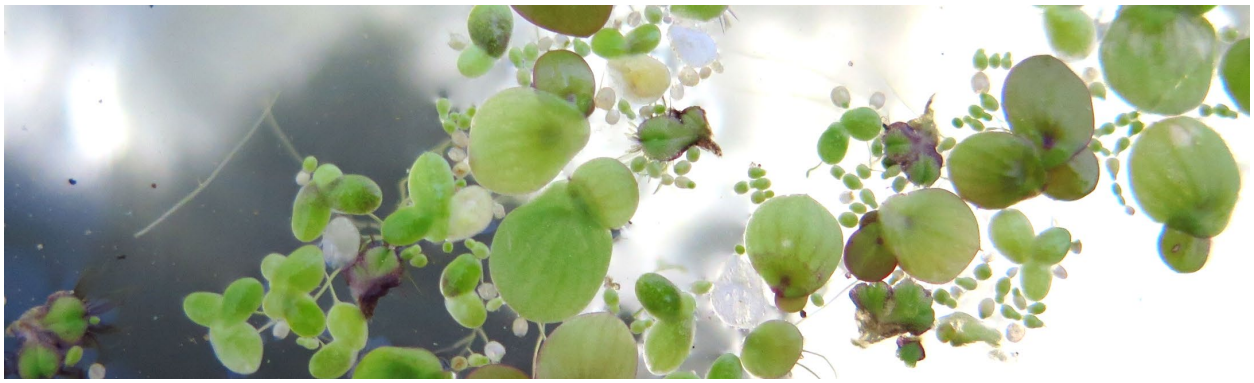
Biodiversity – Ecosystem functioning in duckweed communities



Question

What is the relationship between the biodiversity in community and its productivity?

Summary

The primary objective of this activity is to illustrate the relationship between plant diversity and community productivity. In this lab you will assemble a series of experimental duckweed communities, manipulating species richness. Each of the three species will be grown in monoculture, in every possible two-species combination as well as the full three-species community. All communities will be inoculated with the same total biomass. Mixed-species communities are inoculated with equal biomass of each species where biomass is measured as number of individual fronds multiplied by the species' average frond mass.

This exercise is completed over two laboratory sessions. In the first part you will assemble the experimental communities which will develop in growth chambers for two weeks. In the second part you will measure primary productivity as production of new biomass for each species of each community. This will be done by first sorting the communities into their constituent species and then counting the number of fronds for each species. Finally, to assess phenotypic plasticity in terms of average frond mass, the total biomass for each species for each community is weighed.

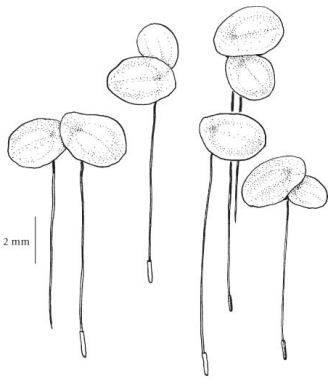
Introduction

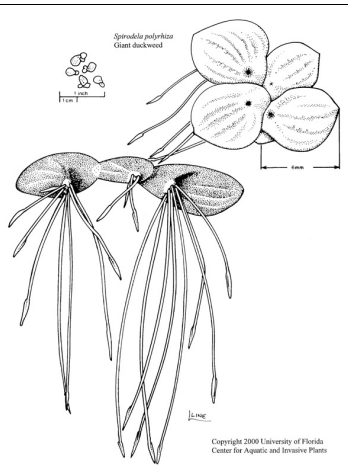
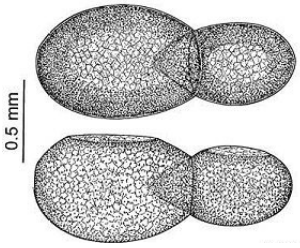
Duckweed (*Lemnaceae*) is a family of small, morphologically reduced floating aquatic monocots. Consisting of five genera and 37 species, they are widespread, growing on every continent except Antarctica. Although reproduction is almost always by asexual and vegetative, certain environmental conditions may lead to the production of flowers and sexual

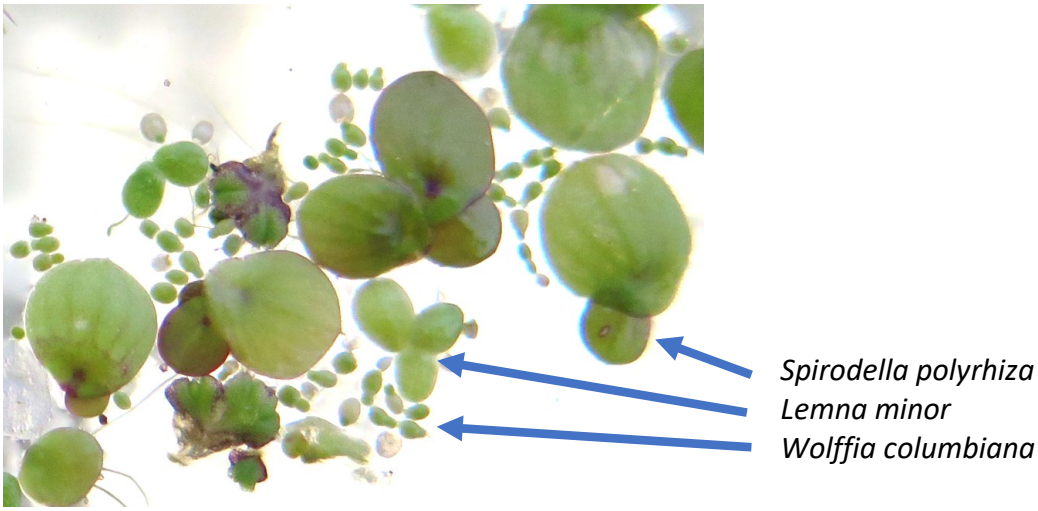
reproduction making them the smallest known flowering plants (Angiosperms). Rapid growth often leads to the formation of clonal mats covering still mesotrophic and eutrophic ponds. Their reduced morphology consists of a single floating frond or thallus and in the case of the genus *Lemna*, a single root, *Spirodela* several roots, or *Wolffia* and *Wolffiella*, no roots.

The last couple decades have seen a rapid growth in duckweed research and application. Two species in particular, *Lemna minor* and *Spirodela polyrhiza* have become model systems in ecotoxicology and are being developed for applications including agricultural and aquaculture animal and fish feed, wastewater remediation and biofuel production. They also serve as a useful model for ecological experiments.

Although the common ducked (*L. minor*) sometimes grows in dense monocultures covering the entire surface of ponds, it is often found in diverse communities, coexisting with other species of duckweed and other floating plants like liverworts. Liverworts are a group of primitive non-vascular seedless plants that reproduce using spores and often resemble mosses, to which they are closely related. Although most species of liverworts are terrestrial, some have reverted to an aquatic life, and some, like *Ricciocarpus spp*. May have both terrestrial and aquatic forms. Although they possess a sexual phase, like duckweed, the vast majority of their reproduction is asexual and vegetative.

Lemna Minor 	**Common name:** Minor duckweed **Description:** Morphology consists of a single frond and single root. The ventral surface is green. Frond diameter between 2-5mm. Daughter and grand-daughter fronds often remain attached to the grandmother to produce clusters of 3-8 individuals.	
Spirodela polyrhiza	**Common name**: Major duckweed **Description:** Morphology consists of a single frond, each with several (between 2-12) roots. The ventral surface is purplish. Daughter and grand-daughter fronds often remain attached	

	to the grandmother to produce clusters of 3-8 individuals. The largest frond diameter of the 3 species, on average 5-10mm.	
Wolffia Columbiana 	**Common name:** Watermeal **Description:** Morphology consists of a single frond with no roots. Fronds measure only ~1 mm, significantly smaller than other species. Mother fronds will produce a single daughter frond and then divide before the daughter frond can produce their own offspring resulting in 2 attached fronds.	



PART 1

Develop your predictions:

1. How might community biomass change as a function of the number of species in the community? Describe both graphically and in writing.
2. What mechanisms might influence productivity in a multi-species community?

Materials (per group of 2 students):

- 1.5L of 10% Hoagland's growth media
- graduated cylinders to dispense media into flasks
- 7 250mL Erlenmeyer flasks
- 2 bacterial loops
- 3 beakers full of each of the 3 species
- labelling tape
- marker

Methods:

Clonal populations of each species have been propagated in the lab under sterile conditions. Given that populations originate from a single individual, intraspecific diversity is negligible, originating only form mutation. Fresh nutrient-rich growth media has been prepared in advance in which to grow the experimental communities.

- In a group of 2, acquire all necessary materials.
- Fill all (7) Erlenmeyer flasks with 150 mL of growth media
- Label the flasks as follows:
- Species richness, Species codes, Group number
  For example,
  1, Lm, 3 indicates *Lemna minor* in monoculture, belonging to group 3
  3, Lm-Sp-Wc, 3 indicates the full 3-speces community, belonging to group 3
- Next, you will inoculate your flasks with the corresponding duckweed species to generate the desired communities. Each flask should start with a total of 150mg of biomass. Using the bacterial loop, hook fronds one at a time, taking care not to break off roots.

Species name	**Species Code**	**Average frond mass**
Lemna minor	Lm	1mg
Spirodela polyrhiza	Sp	4mg
Ricciocarpus natans	Rn	15mg

Calculate the number of fronds for each species to be added to each flask.

Monoculture

Lm: _____
Sp: _____
Rn:_____

2 species communities

Lm: _____
Sp: _____
Rn:_____

3 species community

Lm: _____
Sp: _____
Rn:_____

**A note on frond counting.
Since data will be pooled across groups, it is essential that there is consistency between groups when it comes to frond counting. The simplest standardized protocol is to count all daughter and grand-daughter fronds as individuals, even when still attached. This means that frond count should include all budding fronds visible to the naked eye. For *Ricciocarpus natans*, count each lobe as an individual.

Cultures are then transferred to controlled growth chambers for two weeks at the following conditions: 200umol light /m2/s, light-dark cycle of 16/8, 25^{o}C.

PART 2

Materials (per group of 2):

- the 7 flasks from Lab 1
- 3 beakers
- 3 bacterial loops
- 2 counters
- 1 large tub
- 1 balance
- 1 strainer
- weighing trays
- paper towel
- camera (phone)

Methods:

For each flask:

- Empty the contents into the tub.
- Sort the community by species, isolating each species into its own beaker. Use your clickers to count the number of individuals as you go.
- Record frond number on your data sheet.
- After species have been sorted, counted and recorded for a community, measure the wet mass of each species in the community.
- Strain one species, empty the biomass onto paper towel, blot dry by pressing plants between two sheets (like pressing leaves), then empty contents into a weighing tray.
- Record the total mass for each species for each community on your data sheet.

Data sheet

Species	Species richness	Other species in the community	Number of fronds	Total wet-mass (mg)