Lab 2. POPULATION GROWTH IN *Lemna* (Duckweed)

A. Introduction

An understanding of the dynamics of population growth and the factors that may influence changes in population size is important in many ecological settings. This is particularly true for wildlife and fisheries biologists and plant and animal ecologists interested in protecting endangered species (plants and animals), scientists concerned with biodiversity and habitat preservation, and agroecologists whose interests lie in managed ecosystems.

This lab will give you the opportunity to collect and analyze data on population growth for a rapidly reproducing plant species, *Lemna minor* or duckweed. In addition, you will gain experience in experimentally manipulating populations and learn how density-independent and density-dependent factors affect population growth.

Lemna is one of six genera in the monocot plant family Lemnaceae. All members of this family are small aquatic plants whose dominant mode of reproduction is vegetative (budding). Duckweeds are typically described as free-floating, small (1-2 mm) leafless plants often with a simple, threadlike single root. The small plant body is termed a frond or thallus. These are flowering plants (among the world's smallest) but they rarely flower. Instead, they are capable of very rapid reproduction via the production of buds that break free from the parent plant. Duckweed stands can be very dense in stagnant freshwater achieving densities of 84,000 plants per square meter. Duckweeds are a food source for waterfowl and fish and are dispersed on the feet and feathers of waterfowl.

B. Background

Several mathematical models have been developed to describe population growth and these models vary in complexity, realism, and applicability.

In its simplest form, population growth can be described as:

$$R = B - D$$

where R = rate of growth, dN/dt
B = birth rate,
D = death rate, and

where N = population size, and
t = time,

This equation describes growth of entire populations; however, in some instances it is important to know the rate of change per individual. For example, two populations may have the same population growth rate (change in numbers per unit time), but if the first population is twice as large as the second, the rate of increase (per individual) of the first will be only half that of the second.

If environmental conditions are not limiting such that growth can proceed at its maximum rate, then an exponential growth model describes changes in population size through time (Fig. 2.1). The *discrete exponential equation* for a population of N organisms is:

$$\frac{dN}{dt} = rN$$

where r = intrinsic rate of increase (per individual) or b-d (instantaneous birth and death rates) and
dN = difference between N at time t = n + 1 and N and time t = n.

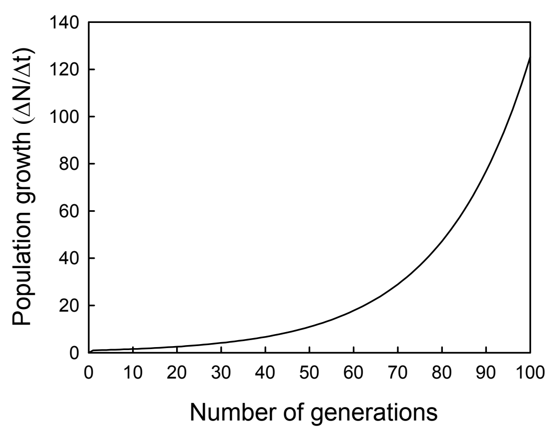
The compound interest analog to this equation is:

$$N_t = N_o * e^{rt}$$

where N_o, N_t = number of individuals at time zero and at time t
e = the base of natural logarithms (2.71828)

The population growth rate (r) is an important species-specific parameter to know. For example, one can calculate a doubling time for a population under non-limiting conditions for growth with an estimate of r ($t = 0.693/r$).

Figure 2.1. An example of exponential growth with r = 0.05 from an initial population of N = 20.



Although examples of exponential growth can be found in natural populations, (either when populations are small and growth is unaffected by limitations due to space, food or species interactions, or when a large proportion of the population is periodically removed), populations cannot grow exponentially for long periods of time.

A more realistic model that incorporates the effects that resource limitations and species interactions may have on population growth is the *logistic model* (Fig. 2.2). The logistic equation for growth of a population with N organisms and a carrying capacity K is:

$$\frac{dN}{dt} = r * N\left(\frac{K-N}{K}\right) \text{ or } \left(\frac{1-N}{K}\right)$$

where r = intrinsic growth rate,
K = carrying capacity (the maximum number of organisms that can be supported indefinitely by a given area or habitat), and
dN = difference between N at time t = n + 1 and N and time t = n.

When $N < K$ the population grows until $K - N = 0$, the population stops increasing. Conversely, if the population were above the carrying capacity (i.e., $K < N$), the population starts to decrease. Figure 1.2 demonstrates the sigmoid curve of the logistic equation.

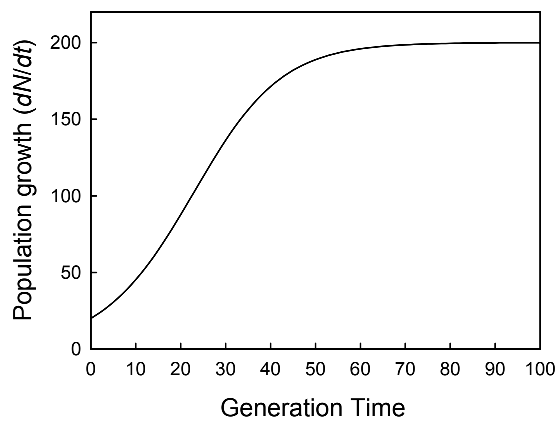


Figure 2.2. An example of logistic growth with r = 0.1, K = 200 from an initial population of N = 20.

Although the logistic population growth model is more realistic than the exponential model, this model also has many assumptions that are often not met by natural populations. These assumptions include:

1) All individuals in the population have identical ecological properties, that is, they have equal probability of producing young, death, and predation.

2) Changes in the population's birth and death rate occur instantaneously, without lag, with changes in population density.

3) All members of the population are equally affected by crowding. This is true only if the population is dispersed uniformly.

4) There is a constant upper limit to population size (K is constant), implying that the environment is constant and the population never increases over K.

5) The population has a stable age distribution.

6) Birth and death rates are not affected by abiotic factors in the environment (density independent events).

Even with these assumptions, some natural populations do conform to logistic growth while others do not. Laboratory organisms with short generation times that are grown under controlled conditions are most likely to behave in the manner predicted by this model.

C. Objectives

For this lab, our objectives are to estimate and compare *r* and *K* for 5 different populations of *Lemna* subjected to different treatments. We will then use the estimates of *r* and *K* to model growth of the populations under different resource availability over a longer period of time.

D. Hypotheses

The following hypotheses will be tested by this study:

1. A logistic growth model will describe population growth in *Lemna* grown under controlled environmental conditions.

2. Estimated values for *r* and *K* will be higher for populations in the resource-rich environment (pond water) compared to the resource-poor (distilled water) environment.

3. Addition of fertilizer will increase estimated *r* and *K* of populations in the resource-poor environment, but not in resource-rich environment.

These hypotheses represent three fundamental steps in basic ecological studies. The first hypothesis relates to our general conceptual understanding of the process or phenomena studied. The second hypothesis relates to how this process varies under natural conditions and includes some assumptions about the reason for this variation. The third hypothesis is the most specific and utilizes an experimental manipulation to help identify the mechanism(s) responsible for the natural variations that may occur under natural conditions.

E. Procedure

Experimental design

Each group (5-6 students each) will grow 10 populations of *Lemna* (2 replicates of 5 treatments), with these populations experiencing four light treatments and one dark treatment. The light treatments are: 1) distilled water only, 2) pond water only, 3) distilled water plus fertilizer 4) pond water plus fertilizer, and the dark treatment is 5) distilled water only (control treatment).

Begin each population with approximately 20 *Lemna* from a stock culture and add them to a cup approximately ¾ full of distilled water or pond water. Label each cup with the group's name and the treatment. **An "individual" will be defined as a frond (or "leaflet") at least 50% as large as mature fronds; there are usually three or four individuals in a cluster**. Not every frond has a root. Do not add white individuals—these are dead plants. If white individuals are attached to green leaflets, simply count the green leaflets. Populations will be grown in small cups.

For the fertilizer treatments, add 2 drops of liquid fertilizer to the distilled and pond water at the beginning of the experiment and every 7 days thereafter. This will ensure that these populations will be continuously fertilized whereas the distilled water control populations should be very nutrient poor.

After the 10 cups for your group are labeled and individuals are counted, place the "dark treatment" cups in a cupboard and the remaining 8 cups under lights on a lab bench.

Measurements

Every two days over the next few weeks each group will census the populations (discard the dead individuals if they are not attached to living plants) and record the number of individuals on a group data sheet as well as an individual data sheet. **Each time you count, refill the cups with distilled or pond water as the water level decreases. Also, make sure to place individuals that are stranded on the sides of the cups back into the water**.

Note: For each population you should have an N_o (for day 0, the day you set up the experiment), and an N_t for t = day 2 and onward. From these data, you will calculate r and K for the different populations. Examples of these calculations follow on pages 7 and 8 of this lab.

Related References

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Hutchinson, G.E. 1978. Introduction to population ecology. Yale University Press, New Haven, Conn.

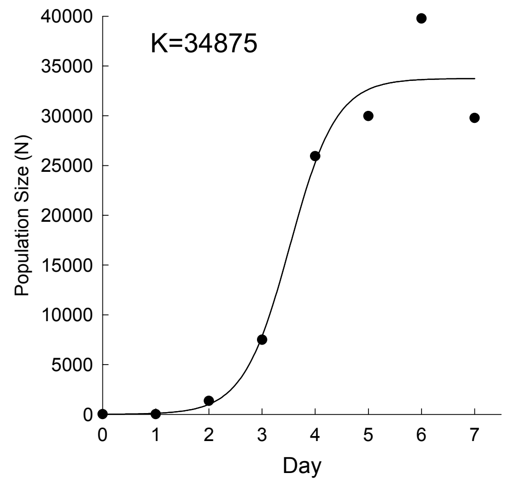
Data Sheet for *Lemna* Population Growth

	Dark Treatment		Light Treatments							
	Control		Distilled		Pond		Distilled +F		Pond +F	
Day	a	b	a	b	a	b	a	b	a	b
0										

Lab 2. Example for Calculating *r* and *K*

1. Graph Population Growth

Day	*N*
0	24
1	22
2	1345
3	7494
4	25940
5	29975
6	39775
7	29775



2. Estimating *K* and *r*

From these data you can calculate *r*, the intrinsic rate of increase, and *K*, the population carrying capacity.

K is estimated by averaging the population size once the population has reached a maximum. In this example, *K* is determined by averaging *N* for days 6 and 7, *K* = 34775.

Calculate *r* mathematically:

The basic logistic population growth equation is:

$$\frac{dN}{dt} = r * N\left(\frac{K - N}{K}\right) \qquad \text{(Eq. 1)}$$

Equation 1 can be rewritten as:

$$r = \left(\frac{dN}{dt}\right) * \frac{1}{N}\left(\frac{K}{K - N}\right) \qquad \text{(Eq. 2)}$$

Equation 2 can be approximated by:

$$r = \left(\frac{\Delta N}{N * \Delta t}\right) * \left(\frac{K}{K - N}\right) \qquad \text{(Eq. 3)}$$

where

ΔN = change in N over a discrete time interval (one, two days)
Δt = time interval (one, two days)
N = population size at the beginning of the time interval

You can use Equation 3 to calculate values for *r* for several values of ΔN and Δt , and thus determine a mean value of *r*.

3. Calculating *r*

Example 1: $\Delta t = 1$ day $K = 34875$ (average of days 6 and 7)

Day	N	ΔN	$\Delta N/N * \Delta t$	$K/K-N$	r
0	24	–	–	–	
1	22	–	–	–	
2	1345	1323	60.14	1.0006	60.2
3	7494	6149	4.57	1.04	4.8
4	25940	18446	2.46	1.27	3.1
5	29975	4035	0.16	3.90	0.6
6	39775	10000	0.33	7.12	2.4
7	29775	-10200	-0.26	–	–
				average	14.21
				w/o 60.2	2.72

Example 2: $\Delta t = 2$ days $K = 34975$ (average of days 6 and 7)

Day	N	ΔN	$\Delta N/N * \Delta t$	$K/K-N$	r
1	22	–	–	–	
3	7494	7472	169.8	1.0006	169.9
2	1345				
4	25940	24595	9.14	1.04	9.5
3	7494				
5	29975	22481	1.50	1.27	1.9
4	25940				
6	39975	14035	0.27	3.87	1.05
5	29975				
7	29975	0	0		
				average	45.6
				w/o 169.9	4.15

Mean value of *r* = (2.72+4.15)/2 = 3.4 or (14.21+45.6)/2 = 29.9

OUTLIERS. Note that in the two sample calculations shown above, two means are shown; one with all the data included and one in which a datum is omitted from the estimate. A scientist must decide whether there is sufficient evidence to exclude a datum that falls well outside the expected range of values. If a datum is thought to be an outlier, either due to gross observer error, problems with equipment, or some natural–caused anomaly that the researcher does not believe represents the system faithfully, the point or points can be removed from analysis. However, it is always important to have a firm reason, methodologically or biologically, for removing any points from your data set. When you estimate *r* and *K* values, you must decide whether or not there are outliers and whether or not to include them in your calculations.