Exercise 3B
Estimating Population Size: Mark-Recapture

Introduction
One of the goals of population ecologists is to explain patterns of species distribution and abundance. In today's lab we will learn some methods for estimating population size and for determining the distribution of organisms.

Measuring Abundance: Mark-Recapture
Mobile animals are usually simpler to define as individuals, but harder to count, because they tend to move around, mix together, and hide from ecologists. Quadrats are not a good approach with mobile animals because immigration and emigration in and out of the study site make it hard to know what area the entire population occupies. For largemouth bass in a farm pond, you could easily draw a line around a map of the population, but how would you define the edges of a population of house sparrows in your community? Although house sparrows tend to be more concentrated in towns and urban areas, they do not stop and turn back at the city limit sign. For zoologists, a fuzzy definition of the space occupied by the population often forces an arbitrary designation of the survey group, such as the "population" of robins nesting on your campus in the spring. Knowing the number of animals in a designated study area is interesting, but we must bear in mind that the ecological population is defined in terms of interactions among organisms of the same species, and not by the ecologist's convenience.

After defining the individual and establishing the limits of the population you wish to count, your next task is to choose a counting method. Arctic and prairie habitats lend themselves to accurate survey by aerial reconnaissance. This approach works poorly in forests, at night, underwater, or in soil habitats. If animals can be collected or observed in a standard time or collecting effort, you can get an idea of relative abundance, but not absolute numbers. For example, the number of grasshoppers collected in 50 swings with an insect net through an old field community produces data that could be used to compare relative abundance in different fields, but would not tell you how many grasshoppers were in the population.

For estimates of absolute numbers, mark-recapture methods can be very effective. The first step is to capture and mark a sample of individuals. Marking methods depend on the species: birds can be banded with a small aluminum ankle bracelet, snails can be marked with waterproof paint on their shells, butterflies can have labels taped to their wings, large mammals can be fitted with collars, fish fins can be notched, and amphibians can have nontoxic dyes injected under the skin. Marked animals are immediately released as close as possible to the collection site. After giving the animals time to recover and to mix randomly with the whole population, the ecologist goes out on a second collecting trip and gathers a second sample of the organisms. The size of the population can then be estimated from the number of marked individuals recaptured on the second day.
The assumption behind mark-recapture methods is that the proportion of marked individuals recaptured in the second sample represents the proportion of marked individuals in the population as a whole. In algebraic terms,

\[ \frac{R}{S} = \frac{M}{N} \]

- \( R \) = animals recaptured on a second day
- \( S \) = size of the sample on the second day
- \( M \) = animals marked and released
- \( N \) = population size

This method is called the Lincoln-Peterson Index of population size.

Let's consider an example. Suppose you want to know how many box turtles are in a wooded park. On the first day, you hunt through the woods and capture 24 turtles. You place a spot of paint on each turtle's shell and release all turtles back where you found them. A week later you return, and with an extraordinary effort, catch 60 turtles. Of these, 15 are marked and 45 are unmarked. Since you know how many turtles you marked, sampled, and recaptured, you can figure out the size of the whole population. By the definitions above, \( M = 24 \) marked and released, \( S = 60 \) in the second sample, and \( R = 15 \) recaptures. If the second sample is representative of the whole population, then:

\[ \frac{15}{60} = \frac{24}{N} \]

This can be rearranged to:

\[ N = \frac{(24)(60)}{15} = 96 \text{ turtles} \]

This method is called the Lincoln-Peterson Index of population size.

In the rearranged version of the general formula, notice that the smaller the number of recaptures, the larger the estimate of population size. This makes good biological sense, because if the population is very large, the marked animals you release into the wild will be mixing with a greater number of unmarked animals, so you will recapture a lower percentage of them in your second sample.
The Lincoln-Peterson method is fairly simple, and its calculations are straightforward, but it does depend on several assumptions. Violating the conditions of the Lincoln-Peterson model can seriously affect the accuracy of your estimate, so it is very important to bear these assumptions in mind as you interpret your results:

1. Individuals with marks have the same probability of survival as other members of the population. It is important to choose a marking method that does not harm your animal. If a predator used your paint marks to locate and capture marked turtles at a higher rate than other turtles, your number of recaptures would be lower, and the estimate would therefore be too high.

2. Births and deaths do not occur in significant numbers between the time of release and the time of recapture. If marked individuals die and are replaced with newborns, then you will recapture few or no marked individuals, and your estimate will be too high. This is not a large concern in studies of box turtles, but can significantly affect estimates for rapidly breeding organisms.

3. Immigration and emigration do not occur in significant numbers between the time of release and the time of recapture. If marked individuals leave the study area and new unmarked individuals come in to replace them, you will get fewer recaptures than the equilibrium population size would lead you to expect. To think about this another way, the real population covers a much larger area than the habitat you thought you were studying.

4. Marked individuals mix randomly with the population at large. If your marked turtles do not move among unmarked turtles, and you recapture them near the place you released them, then recaptured turtles may be overrepresented in your second sample, driving down your population estimate.

5. Marked animals are neither easier, nor harder, to capture a second time. If marking an animal frightens it so that it hides from you a second time, then recaptures will be underrepresented in a second sample. If animals become tame and are easier to recapture, then the opposite error is introduced.

6. Marks do not come off of your marked organisms. Invertebrates molt and shed marks, mammals can wriggle out of their collars, and many things can happen to obscure your marks. If this happens, recaptures will be undercounted, and your estimate will be too high.
7. Recapture rates are high enough to support an accurate estimate. The Lincoln-Peterson calculation tends to overestimate the population size, especially if the number of recaptures is small.

Assumption 7 is often violated, because it is difficult to generate sufficient recaptures in large populations. To provide a better estimate, ecologists sometimes use multiple marks and recaptures and the **Schnabel index** to estimate population size:

\[
N = \sum_{i=1}^{m} \frac{M_i C_i}{R_i}
\]

where \(M_i\) = the total number of previously marked animals at time \(i\), \(C_i\) = the number caught at time \(i\), and \(R_i\) = the number of marked animals caught at time \(i\).

As an example, suppose we caught and marked 100 animals in our first sample, captured 85 animals (15 marked and 70 unmarked) in a second sample, and then captured 105 animals (25 marked and 80 unmarked) in a third sample. We would then have the following:

**Table 3.1** Example of Schnabel Index Calculation

<table>
<thead>
<tr>
<th>(i)</th>
<th>(C_i)</th>
<th>(R_i)</th>
<th>New marked</th>
<th>(M_i)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>100</td>
<td>0</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>85</td>
<td>15</td>
<td>70</td>
<td>100</td>
</tr>
<tr>
<td>3</td>
<td>105</td>
<td>25</td>
<td>-</td>
<td>170</td>
</tr>
</tbody>
</table>

Note that \(M_i\) at time 1 is zero; this happens because we start this sampling with no marked animals. For each subsequent time period, \(M_i\) is simply the sum of all previous values in the "New marked" column. From the above, we can estimate population size as:

\[
N = \frac{(0*100) + (100*85) + (170*105)}{0 + 15 + 25} = \frac{26350}{40} = 659
\]

Note that had we sampled only two times, our population size estimate would be 567, so this value is dependent on the number of samples taken.
Lab Exercise 3B: Mark-Recapture of Pinto Beans

Research Question
How do population estimates using mark-recapture techniques compare to the true population size?

Preparation
Mark-recapture involves marking a set number of organisms, releasing them back into the population where they mix with unmarked individuals, and then doing a second collecting visit. Since mark-recapture typically requires a substantial time between marking and recapturing (at least 24 hours, and generally longer), we will simulate the process using the incredibly non-mobile pinto bean in place of a mobile animal.

Materials (per laboratory team)
Container to hold 400 beans
400 beans
Sampling container (such as a centrifuge tube)
Marker

Procedure
1. Obtain a container and add 400 beans to it.

2. Obtain a sampler (a centrifuge tube) and collect a sample of beans by filling the sampler to the top. Count and record the number collected and mark each collected bean with a marker. After marking, return the beans to the population and thoroughly mix the beans by shaking the container.

3. Draw out a second sample. Record the number of marked and unmarked beans, but do not return this to the population just yet. You will use these numbers to estimate population size using the Lincoln-Peterson index.

4. Now mark each unmarked bean and return this sample to the population, thoroughly mix, and then take a third sample. Again, count the number of marked and unmarked beans, mark the unmarked beans and return the sample to the bean population.

5. Take a fourth and final sample, and count marked and unmarked beans. Now you have the data necessary to estimate population size using the Schnabel index.
Data Analysis: Population Size Estimate
1. Enter your data in the table below.

Table 3.3 Mark-Recapture Data

<table>
<thead>
<tr>
<th>i</th>
<th>$C_i$</th>
<th>$R_i$</th>
<th>New marked</th>
<th>$M_i$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
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<tr>
<td>2</td>
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<td>3</td>
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<tr>
<td>4</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

2. Using the Lincoln-Peterson index, calculate the number of pinto beans in the population (N). Show your calculations.

Lincoln-Peterson Index:

$$N = \frac{M \cdot S}{R}$$

N = population size estimate
M = marked individuals released
S = size of second sample
R = marked animals recaptured

3. Using the Schnabel index, calculate the number of pinto beans in the population (N). Show your calculations.

$$N = \frac{\sum_{i=1}^{m} M_i C_i}{\sum_{i=1}^{m} R_i}$$

Discussion
How do your estimates compare to the true population size? If your estimated values differ from the true value, why do you think this might have occurred?

Did the Schnabel index give you a better estimate of the actual population size than did the Lincoln-Peterson index? Why?