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Ants as Model Organisms to Study Species Coexistence

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Community ecologists seek to understand mechanisms regulating species coexistence. Decades of experimental research suggest that niche differentiation plays an important role in allowing ecologically similar species to coexist. In this lab, we will perform a field experiment to test for niche differentiation in an ant (Hymenoptera: Formicidae) community. We will have four main objectives: ***1)*** appreciate ant diversity, ***2)*** test hypotheses about how niche differentiation mediates species coexistence, ***3)*** use ecological methods and analyze data, and ***4)*** participate in School of Ants, a global citizen science initiative based in North Carolina that uses ants to explore biological diversity.

Keywords: ants, community ecology, niche partitioning, interspecific competition, species coexistence

Introduction

This lab is intended to introduce undergraduate students to basic concepts in community ecology. The methods are simple (*e.g.* involve placing baits on notecards), and the lab can be adjusted for any experience level. To increase rigor for upper level students, more sophisticated statistical analyses can be added. For entomology students, a greater emphasis can be placed on species identifications. This lab can also be geared towards biology majors or non-majors. For most students, distinctions based on morphological traits (i.e. morphospecies identifications) will suffice. Even if the students have little or no experience with insects, they will be able to distinguish between large slow black ants and fast small yellow ants. This is because each student will likely only encounter at most four ant species foraging on a given notecard. This lab can be completed in a single three-hour lab period. The fieldwork can take less than an hour, from establishing transects to cleaning up the notecards. This lab can be used to introduce students to fieldwork and ecological sampling techniques, as well as animal ecology and behavior.

Student Outline

Ants as model organisms to study species coexistence

Make sure to bring:

Notebooks, writing utensils, field clothes (hat, long pants, closed-toe shoes), sunscreen, water bottles.

Main question:

How do groups of species with similar ecological needs coexist in environments with limited resources?

Goals:

1) Learn to observe ant communities
2) Test hypotheses central to the field of community ecology
3) Implement ecological sampling techniques and analyze resulting data
4) Contribute to a national database of urban ant diversity maintained by the School of Ants (**www.schoolofants.org**)

Key concepts:

Ecological niche— A summary of an organism's tolerances, requirements, and behavior. Note that the niche is not a tangible thing or place.
Interspecific competition—Individuals of one species suffer a reduction in reproductive success, survivorship, or growth as a result of the exploitation of resources or interference by individuals of another species
Competitive exclusion—One species holds a resource at a level that is too low for effective usage by another species
Habitat heterogeneity—The level of variability in environmental conditions at a single locality

Ant Communities: A case study in niche differentiation

Competition for a limited pool of resources (e.g. light, water, food, shelter) influences both how species evolve and where they occur. Niche differentiation is a mechanism allowing ecologically similar species to coexist in a given habitat. For example, two closely related ant species may depend on the same tree species for food, but they may specialize on different seed sizes or forage at different times of the day. More heterogeneous habitats are predicted to support more species because they meet a broader range of niche requirements.

Ants (Hymenoptera: Formicidae) are abundant predators, soil turners, and seed dispersers that dominate terrestrial ecosystems across the planet. Individual ants belong to colonies in which a queen lays eggs and sterile workers divide the tasks of foraging for food, caring for developing nestmates, and defending and building the nest. It is the foraging ants that are conspicuous above ground in their quest to harvest resources. Biologists working in habitats from tropical rainforests to deserts to boreal forests have long used the traits of foraging ants to study whether competition structures communities. Research opportunities are plentiful, given that over 14,000 ant species exhibit a wide range of foraging strategies and resource preferences. A typical North Carolina ant community might have over 60 species!

When observing ants, note that some species are ubiquitous while some are rare, and that some forage in frantic masses while others approach baits slowly and discreetly. Note that some ants follow orderly pheromone-laden foraging trails while others arrive haphazardly. Note that small ants, through sheer numbers and chemical weapons, can displace larger lumbering ant species.

Today, we will test two hypotheses that may explain how niche differentiation may allow ant species to co-exist in a community:

1. ***The Resource Specificity Hypothesis:*** Ant species may coexist through dietary specialization, by subdividing a single habitat. One prediction is that we will find greater numbers of certain species of ants at nutritionally distinct foods. In addition, we will examine foraging of omnivores at baits combining protein and sugar (pecan cookies).
2. ***The Habitat Specificity Hypothesis:*** Ant species avoid competitive interactions by specializing in different habitats. If this is so, one would predict that more heterogeneous habitats should support more species. We will compare the ant communities on pavement or along trails with adjacent vegetated areas (e.g. a forest, or grassy lawn).

Methods

Four groups, each of which will observe five stations, will independently test hypothesis 1. We will evaluate hypothesis 2 as a class after combining each group's data. Data sheets are attached at the end of this handout.
In the field, each group will:

1) Place a transect, either along a trail/on pavement or in the surrounding vegetated areas. Record general microhabitat conditions (i.e. under large juniper bush, or along sandy trail).
2) Measure a 20 m transect, placing three note cards at 5 m intervals. Two students will be responsible for monitoring each set of three note cards. Notecards should be separated by at least 2 m.
3) Crumble a nut-based shortbread cookie (we recommend Keebler Pecan Sandies) on one note card, place a small spoonful of jelly on the second card, and place a spoonful of tuna on the third card.
4) Record the following information at 5 minute intervals (45 minutes total), using the formatted data sheet appended below (Table 1):
    a. Number and identity of ant morphospecies on note cards.
    b. Abundance of each morphospecies (e.g. 1-5, 5-20, 20-40, >40 individuals). Note: count only those ants that are *actually ON* each note card.
5) Record behavioral observations (e.g. large black ant chased away small yellow ant)
6) Prepare ant samples from cookie bait to be sent to School of Ants. See the website **www.schoolofants.org** for instructions.

Homework: Individual Abstract

Use your group's data to evaluate hypothesis 1. Include a graph showing whether the average number of species differed between baits with jelly, tuna, and cookies (*Resource Specificity Hypothesis*). Use the combined class data (your instructor will email the data once they are compiled) to evaluate hypothesis 2: testing whether species richness differed between trail/pavement and vegetated habitats (*Habitat Specificity Hypothesis*). Think about the independent and dependent variables, whether these data are discrete or continuous and thus, what kind of graphs to make. Include a table showing your raw data with all behavioral observations. If desired, two way ANOVAs with 'morphospecies ID' and 'bait type' or 'habitat type' can be used to test hypotheses about the abundance of ant species or total ants at baits. For a sample test of the *Resource Specificity Hypothesis* using data collected when we ran this lab with undergraduate students, see Figure 1 and Table 2.

Your discussion should evaluate the evidence for each hypothesis. Consider these questions:

1) Does species richness differ with bait type, habitat type, or over time?
2) How do species differ in foraging behaviors?
3) What other factors (i.e. experimental error or unmeasured environmental variables) might have affected the results?
4) How might you modify the experiment to better answer the research question?
5) What are possible future directions of this research? What is the broader importance?

Table 1. Data sheet

Data Sheet: NAME_________________________

Time (min.)	**Bait Type** **jelly/tuna/pecan sandie**	**# Morphospp.**	**# of each Morphospp.**	**Behavioral Observations**

Materials

Students supply

Notebooks, writing utensils, field clothes (hat, long pants, closed-toe shoes), sunscreen, water bottles.

We supply (per 25 students)

4 measuring tapes (20-m)
200 3 x 5 notecards
2 bags of pecan shortbread cookies (e.g. Keebler Pecan Sandies)
4 jars of grape jelly
8 cans of tuna (preferably packed in oil)
12 spoons
4 stopwatches
8 hand lenses
1 box of ca. 50 ziploc bags

Notes for the Instructor

General Comments

1. It is generally a good idea to scope out the field site ahead of time, to locate areas of high ant activity, make sure there are no fire ants, and get a feel for the resident ant fauna and the time needed for discovery of pilot baits.
2. Species level identification is not necessary for this experiment. Students just need to distinguish between the various types of ants they see, and to standardize the names as a class (e.g. large black, slow yellow, fast red). This is feasible because each pair of students will be in charge of observing three notecards (jelly, tuna, cookie) and will probably encounter at most three or four ant morphospecies. For instance, they will be able to distinguish between slow moving yellow ants vs. large fast black ants. Students will rapidly gain the confidence and skills to do this; instructors can run between transects to help keep students focused.
3. For identification of ant genera, we recommend: Fisher and Cover (2007) *Ants of North America: A Guide to the Genera.* If desired, instructors can collect local ants, place them in small containers, and allow students to practice differentiating morphospecies before going to the field.
4. This experiment is best run in the early afternoon at temperatures above 20˚C. It will not work in rainy weather.
5. Only ants captured at cookie baits should be sent to the School of Ants.
6. Most ants will not pose a health risk to students. However, some ants can sting (e.g. fire ants).

Sample Results

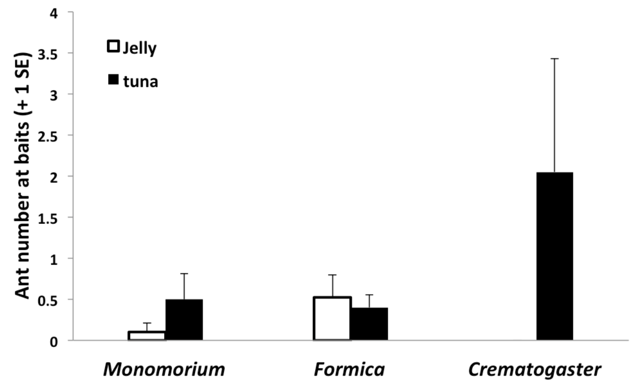

Figure 1. We found mixed support for the *Resource Specificity Hypothesis*. While species tended to be more abundant at a specific bait type, this effect was not statistically significant ($F_{1,111} = 2.42$; $p = 0.12$). This trend was driven by higher abundance of *Crematogaster* at tuna baits relative to jelly, which suggests it was a resource specialist. For this analysis, we used only data at the 25-minute sampling interval.

Table 2. Results of a two way ANOVA testing for species differences across bait types.

Factor	df	Type III SS	F	p > F
Bait type	1	17.46	2.42	0.12
Morpho-species ID	2	5.61	0.78	0.46
Bait x Morpho-species	2	12.58	1.75	0.18
Error	111			

Description of How to Present

Schedule

1:30-2:00 Lecture in the classroom (a brief introduction to ants and ecological field research)
2:00-2:30 Set up experiment, demonstrate techniques
2:30-3:30 Run experiment
3:30-4:20 Analyze data as a class

Methods

Four groups of 10 individuals will independently test hypothesis 1. We will evaluate hypothesis 2 as a class after combining each group's data. A sample datasheet is provided in attached in Table 1, above.

In the field, each group will:

1) Two groups will set up transects along the trails or on pavement and two will set up transects in the surrounding vegetated areas. Record general microhabitat conditions (i.e. "under large juniper bush," or "along sandy trail").

2) Measure a 20 meter transect, placing three note cards at 5 m intervals. Two group members will be responsible for monitoring each set of three note cards. Note cards should be separated by at least 2 meters.

3) Crumble a nut-based shortbread cookie (we recommend Keebler Pecan Sandies) on one note card, place a small spoonful of jelly on the second card, and place a spoonful of tuna on the third card.

4) For 45 minutes, all group members record the following information at 5-minute intervals, using the formatted data sheet appended above in Table 1:

    a. Number and identity of ant morphospecies on note cards.

    b. Abundance of each morphospecies (ex. 1-5, 5-20, 20-40, >40 individuals). Note: count only those ants that are *actually ON* each note card.

7) Record behavioral observations (e.g. large black ant chased away small yellow ant)

8) Prepare ant samples from cookie bait to be sent to School of Ants. See the website **www.schoolofants.org** for instructions.

Literature Cited

Fisher, B. L., and Cover S. P. 2007. A*nts of North America: A Guide to the Genera*. University of California Press, 216 pages.

About the Authors

Dr. Jonathan Shik is a postdoctoral researcher at North Carolina State University. He received a B.Sc. in Biology from McGill University in 2003 and a PhD in Ecology and Evolutionary Biology from the University of Oklahoma in 2010. His dissertation research focused on the evolution and ecology of ant societies with much of the fieldwork performed in a Panamanian tropical rainforest. His current research focuses on how invasive ant species gain a foothold in introduced habitats by engaging in mutualisms with plant sucking insects.

Lauren Nichols is a research assistant at North Carolina State University. Her M.A. research at Wesleyan University focused on the evolutionary ecology of invasive species. In addition to her current research on the impacts of global warming on the ant communities within Eastern deciduous forests, she also assists with the School of Ants project.

Dr. Andrea Lucky is a research scientist in the department of Entomology and Nematology at the University of Florida. Her PhD research at the University of California at Davis focused on the evolutionary biology, taxonomy and conservation of ants in Australia and Melanesia. Currently, her research continues abroad as well as close to home, where she is founding director of the School of Ants project (www.schoolofants.org), a study of urban ant biodiversity that uses citizen science data to document and monitor changing communities.

Dr. Mariëlle Hoefnagels is an associate professor at the University of Oklahoma in the departments of Biology and Microbiology/Plant Biology. She earned a B. Sc. in environmental science from the University of California at Riverside in 1987, an M. S. in soil science from North Carolina State University in 1991, and a Ph.D. in botany and plant pathology from Oregon State University in 1997. Her primary responsibility at OU is to teach nonmajors biology. She is also the author of two university-level textbooks published by McGraw-Hill (*Biology: Concepts and Investigations* and *Biology: The Essentials*).

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