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Teaching Issues and Experiments in Ecology - Volume 7, July 2011

EXPERIMENTS

Are males cheaper than females? Male and female costs of reproduction

Ivana Stehlik

Department of Biological Sciences, University of Toronto at Scarborough, 1265 Military Trail, Toronto, ON, Canada, M1C 1A4
ivana.stehlik@utoronto.ca

ABSTRACT
In this one to two week field project, students investigate the costs of reproduction. In dioecious plants, a female's investment in reproduction is typically much greater than a male's, because while both sexes encounter the basic cost to produce a flower, only females have to allocate energy to seeds, exceeding the energy requirements to produce pollen. This field project tests whether the effects of these unequal costs are reflected in characteristics of individuals of functionally dioecious and long-lived Jack-in-the-pulpit (*Arisaema triphyllum*) in the field. Students will read two introductory articles and take a pre-project online quiz, collect the data in the field, analyze it, collect and interpret literature sources and will write a short report.

KEYWORD DESCRIPTORS
- **Ecological Topic Keywords:** plant ecology, cost of reproduction, sex change, size class, forest herb, dioecy, sequential hermaphroditism, seed production
- **Science Methodological Skills Keywords:** collecting and presenting data, data analysis, evaluating alternative hypotheses, field work, graphing, scientific writing, statistics, use of primary literature
- **Pedagogical Methods Keywords:** formative evaluation, guided inquiry, inquiry-based learning, self-guided reading, cooperative group work, scientific writing skills, one-minute papers

CLASS TIME

One week or 3 lab hours (in addition to any travel time), if instructor identifies where populations of the study plant are and directs students to them.

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OUTSIDE OF CLASS TIME

Six - seven hours to read two introductory articles and take a pre-project online quiz, analyze the data, collect and interpret literature sources and to write a short report.

STUDENT PRODUCTS

- Online quiz as a motivation for self-directed learning and preparation
- Class data set
- Short report (short report such as the brief communications found in many journals, in which students present a short but complete report on the results of the statistical tests that they will perform).

SETTING

This field project is ideally done during the flowering time of Jack-in-the-pulpit (*Arisaema triphyllum*) which is late spring, but because universities hardly ever schedule classes during this time, I have developed this project to be done starting in summer (after the development of seeds; i.e. during summer (field) ecology courses) into early fall (mid to late September), which makes this lab attractive as an activity for fall courses. Jack-in-the-pulpit is a species common to (moist) deciduous forests in Eastern North America, from Florida to Texas in the south to Nova Scotia, Prince Edward Island to Manitoba (USDA, 2009). It is also easy to recognize and spot based on its characteristic three-parted and fairly large leaves.

COURSE CONTEXT

This field activity could be used in two ways: (1) in a third-year ecology course for 12-16 undergraduate students using the instructions as outlined in the present document, where students cooperatively collect their data in groups of two, and (2) in third to fourth year, two-week summer field course setting as an individual student project. In the latter setting, the project approach could be more inquiry-based, as described under **Comments on Translating the Activity to Other Institutional Scales or Locations**.

INSTITUTION

Public research and undergraduate university of approximately 10,000 students.

TRANSFERABILITY

This project would fit general ecology or upper division ecology courses at institutions of all sizes. The lab is simple in design and requires no special technical skills or tools and thus could be transferred to non-majors general biology classes, but access to suitable forested field sites is key.

This project could be run using other (perennial) dioecious species, after a careful prior evaluation by the instructor. Jack-in-the-pulpit offers the added twist of potential inter-annual gender changes (as a sequential hermaphrodite), but the basic questions could be tested with any genetically fixed dioecious plant species.

ACKNOWLEDGEMENTS

This field project has a large body of literature backing it up (e.g., Bierzychudek 1982; Doust and Cavers 1982; Policansky 1987; Vitt et al. 2003). As a course unit, I formulated and optimized this field project while teaching a summer field course in experimental ecology and evolution at the University of Toronto, Koffler Scientific Reserve, as a safe and efficient backup project for failing projects due to adverse weather conditions and running out of time to sample. I especially would like to acknowledge the efforts of Katie Krelove who was the first to conduct this field project and smoothed out some of its initial edges.

SYNOPSIS OF THE EXPERIMENT

Principal Ecological Question Addressed

Sexual reproduction is costly, however, the costs of male versus female reproduction are unequal in most organisms, both plants and animals. In dioecious plants (male-only or female-only individuals), a female's investment into reproduction is typically much greater than a male's over the course of the growing season, because while both sexes encounter the basic cost to produce a flower, only females have to allocate nutrients and energy into seeds, mostly exceeding the biomass and energy requirements to produce pollen. This field project tests whether the effects of these unequal and gender-based costs are reflected in characteristics of individuals of functionally dioecious and long-lived Jack-in-the-pulpit (*Arisaema triphyllum*) in the field. At early life stages, an individual Jack-in-the-pulpit does not reproduce, then will turn male in later years and only later in life reproduce as a female (sequential hermaphroditism). The particular hypothesis tested in this project is whether asexual individuals have the smallest, males intermediate-sized, and females the largest leaves.

What Happens

Students measure leaf length of non-reproductive, male and female individuals of Jack-in-the-pulpit, analyze the data, and prepare a short report on their findings.

To promote self-directed learning, an (online) quiz is implemented to motivate the preparatory readings of two background papers.

Experiment Objectives

- Students will learn to ask questions that generate testable hypotheses about the cost of reproduction, gain experience designing field projects to test those hypotheses, and analyze and present results in scientific format.
- Students will learn to identify Jack-in-the-pulpit and its preferred habitat in a natural setting and find their way around in a forest.
- Students will learn to effectively communicate the purpose, results, and conclusions of this study by writing a short report.

Equipment/ Logistics Required

30 cm ruler, (Rite-in-the-Rain) notebook, pencil

Summary of What is Due

Prior to class, each student will have to read the project instructions, two preparatory background papers and take an online short-answer quiz worth a fraction of the grade.

After field work, each student or group of students must prepare and submit a spreadsheet that contains the collected raw field data which then will be assembled as the class data. This task may be completed upon return to the lab if time permits; if not completed in the lab, students must e-mail or upload the spreadsheet with their data to the instructor, TA and to other students in the class.

Students will produce a short report which includes all sections of a traditional lab report except no abstract is expected. Students must include a figure and a table of statistical results for each test they perform.

DETAILED DESCRIPTION OF THE EXPERIMENT

Introduction

Plant sexual reproduction is costly in terms of the resources required for flowering and fruit set. However, the costs of male versus female reproduction are, in most organisms (including animals), unequal (Lloyd and Webb 1977; Popp and Reinartz 1988). In dioecious plants (male-only or female-only individuals), a female's investment into reproduction is typically much greater than a male's over the course of the growing season (Queenborough et al. 2007). Both sexes encounter the basic cost to produce a flower, but whereas costs for reproduction end after the production of pollen in males, females have to allocate nutrients and energy into seeds, thereby exceeding the biomass and energy requirements to produce pollen by males. This greater resource allocation into reproduction as opposed to vegetative growth (roots, shoots and leaves) in females is reflected in the often observed reduction in vegetative growth of females as compared to males. Thus males can, after the end of flowering, allocate incoming assimilated sugars into growth and defense, whereas females have to continue to allocate resources into their offspring. This tradeoff between growth/defense and reproduction is often reflected in greater height and/or larger annual tree rings in males as compared to females (Vasiliauskas and Aarssen 1992; Cipollini and Whigham 1994; Obeso 1997; Queenborough et al. 2007). Females have also been observed to start flowering later in their life, taking longer to build up the necessary reserves for their more expensive reproduction as compared to males (Bullock and Bawa 1981; Garcia and Antor 1995). In extreme, but not uncommon cases, this higher female allocation into reproduction can lead to higher female mortality (Allen and Antos 1993; Matsui 1995) and hence male-biased population sex ratios (Lloyd and Webb 1977). This is due to the fact that males can afford a costlier defense against herbivores or be better prepared to cope with environmental stress such as drought.

Some species of plants and animals demonstrate the ability to change sex throughout their lives. This sex change is in agreement with the size-advantage model (Warner 1988). The model predicts that an individual should change sex if it can increase its reproduction by doing so. Thus, natural selection will favor sex change in a species if there is differing reproductive success between males and females at different sizes (Charnov 1982; Ghiselin 1969). Policansky (1987) puts size advantage in terms of cost of reproduction, stating that larger individuals are better at bearing the costs of reproduction than smaller individuals.

Jack-in-the-pulpit (*Arisaema triphyllum*) is a long-lived understory herb and a great test case for the size-advantage model for species with labile sex expression. In any given year, a Jack-in-the-pulpit plant is either asexual, male, or female. From year to year, however, an individual has the ability to change its sex expression among all three of those sexual states. As a perennial, excess energy acquired in the past growing season is saved in an underground corm (storage organ) from which the individual regrows in the consecutive spring. This amount of stored energy affects which sexual state a given Jack-in-the-pulpit will express. Males and females face very different expenses for sexual reproduction, as only females grow a large infructescence (i.e. the fruiting stage of an inflorescence) with dozens of fleshy red berries (Fig. 1). The annual amount of stored energy is dictated by the leaf area of the plant, with larger leaves having a greater photosynthetic surface and thus higher capacity for energy production.

Thus, given the fact that (1) reproduction in Jack-in-the-pulpit is generally costly, that (2) females and males face different costs of reproduction and that (3) genders are not fixed, what predictions would you make for the size of leaves in a given plant, considering that leaf area is a correlated with photosynthetic output and hence how much energy is allocated to reproduction? Or put more simply, which sexual state (non-reproductive, female, male) would have either small, intermediate or large leaves? And how would you go about testing your prediction?

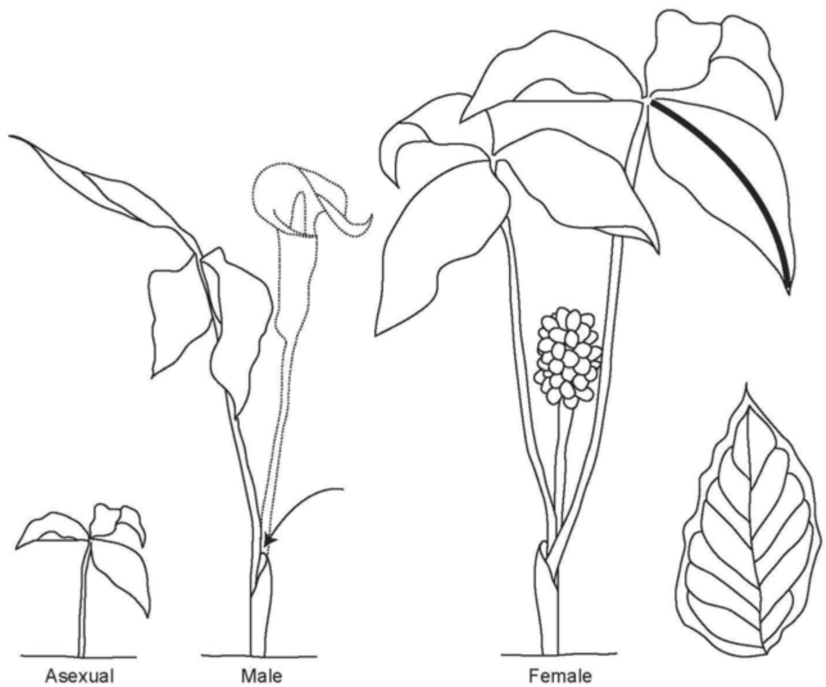

Figure 1. Drawings of Jack-in-the-pulpit (*Arisaema triphyllum*) in the field in late summer to early fall, i.e., when flowering is over. Asexual plants only produce one, typically small leaf (left). Males are typically larger and have a sheath with a lateral hole where previously the male inflorescence (now decayed) was inserted (middle; arrow). Females typically are the largest individuals with typically two leaves and a centrally inserted infructescence with many bright, shiny and red berries (right). Note that some males have two leaves and, in such a case, the hole left behind by the withered inflorescence would be central between the two leaves, such as in the case of females (but obviously with no evidence of an infructescence). The thick line on one of the female leaves depicts how students should measure the length of the largest leaflet of the largest leaf. The single leaflet on the far right demonstrates the characteristic venation of Jack-in-the-pulpit, i.e. with few very conspicuous large veins on either side of the leaf joining some distance of the leaf margin. Drawings: Ivana Stehlik.

Materials and Methods

Study Site(s)

Jack-in-the-pulpit (*Arisaema triphyllum*) is a species common to (moist) deciduous forests in Eastern North America, from Florida to Texas in the south to Nova Scotia, Prince Edward Island to Manitoba (USDA, 2009).

Overview of Data Collection and Analysis Methods

1. Data collection

Together with your instructor, decide whether you will work in groups of two or alone. Once the outdoor data are collected, each student or student group contributes and obtains access to the whole class data set for use in the individual analysis and write-up.

As a whole class, collect data on approximately 50 - 100 sexable plants, thus calculate how many plants a group or individual student needs to assess. However, because of the typical prevalence of asexual plants and the, relatively speaking, smallest fraction of females in natural populations of Jack-in-the-pulpit, students should measure a required minimum number of males and females. Based on experience and class sizes of 12 - 16, three males and females per person (or six females and males per group of two students), respectively, and more easily obtainable ten (20) asexuals should create a strong enough class data set.

2. Field work

2.1. Leaf size measurement

Measure non-destructively the length of the longest leaflet of the largest leaf (Fig. 1, thick line). Leaf length is a strong predictor for the overall leaf area, hence field work can be much simplified by only assessing one parameter per plant individual. Based on previous experience, the following equation

(leaf length x leaf width)/ 3.17

is a strong predictor of actual leaf area ($R^2 > 0.8$).

2.2. Sex expression

In the field and without removing any Jack-in-the-pulpit individuals, students should determine the functional gender of each plant by first looking for the presence of an infructescence (stalk with berries; Fig. 1) and where one is found, the individual plant should be recorded as female. In the absence of an infructescence, students should search for evidence of a withered (male) inflorescence, i.e. a hole near the base of a leaf stem (Fig. 1; arrow for the location of a potential hole). If such a hole can be found, the individual should be recorded as male. Where neither an infructescense nor evidence of (a withered) inflorescence is found, the individual should be recorded as asexual.

3. Data analysis
Using the class data file, specify "leaf length" as continuous and "sex" (A: asexual; M: male; F: female) as categorical. Based on the level of stats aimed for, run a box-plot analysis or one-way ANOVA (with post-hoc tests [e.g. Tukey-Kramer or Bonferroni] to test for pair-wise differences between genders). From the analysis, retrieve the mean leaf length per functional gender including standard errors or confidence intervals.>

Questions for Further Thought and Discussion

Potential questions for student discussions or for one-minute papers:
Questions 1 – 3 can be discussed before the students head out into the field, whereas questions 4 and 5 should follow the data processing, but possibly before the write-up.

(1) Some females might not receive any pollen and hence their inflorescences might wither, similarly to those of males. Such individuals would be scored as males. How does that influence the data set?
(2) What happens to a female Jack-in-the-pulpit if it were to suffer herbivory e.g. by deer?
(3) How might abiotic limitations (such as light) influence sex expression in Jack-in-the-pulpit? What could be measured to assess this?
(4) In the introduction, you have learned that in some dioecious species (where individuals are fixed as either male or female), males are taller and more vigorous. Yet in Jack-in-the-pulpit, females are expected to be taller than males. Why?
(5) If it is cheaper to be a male and each plant still tries to best defend itself against the perils of its biotic (herbivory) and abiotic (e.g. drought) environment, why don't all plants 'choose' to reproduce as males and hence increase their chance for survival?

References and Additional Resources

Allen, G. A. and J. A. Antos. 1993. Sex ratio variation in the dioecious shrub *Oemleria cerasiformis*. American Naturalist 141: 537-553.
Bierzychudek, P. 1982. The demography of jack-in-the-pulpit, a forest perennial that changes sex. Ecological Monographs 52: 335-351.

Bullock, S. H. and K. S. Bawa. 1981. Sexual dimorphism and the annual flowering pattern in *Jacaratia dolichaula* (D. Smith) Woodson (Caricaceae) in a Costa Rican Rain forest. Ecology 62: 1494-1504.

Charnov, E. L. 1982. The Theory of Sex Allocation. Princeton University Press, Princeton, New Jersey, USA.

Cipollini, M. L. and D. F. Whigham. 1994. Sexual dimorphism and cost of reproduction in the dioecious shrub *Lindera benzoin* (Lauraceae). American Journal of Botany 81: 65-75.

Doust, J. L. and P. B. Cavers. 1982. Sex and gender dynamics in Jack-in-the-pulpit, Arisaema triphyllum (Araceae). Ecology 63: 797-808.

Garcia, M. B. and R. J. Antor. 1995. Sex ratio and sexual dimorphism in the dioecious *Borderea pyrenaica* (Dioscoreaceae). Oecologia 101: 59-67.

Ghiselin, M. T. 1969. The evolution of hermaphroditism among animals. Quaterly Review of Biology 44: 189-208.

Lloyd, D. G. and C. J. Webb. 1977. Secondary sex characters in plants. Botanical Review 43: 177-216.

Matsui, K. 1995. Sex expression, sex change and fruiting habit in an *Acer rufinerve* population. Ecological Research 10: 65-74.

Obeso, J. R. 1997. Costs of reproduction in *Ilex aquifolium*: effects at tree, branch and leaf levels. Journal of Ecology 85: 159-166.

Policansky, D. 1987. Sex choice and reproductive costs in jack-in-the-pulpit: size determines a plant's sexual state. Bioscience 37: 476-481.

Popp, J. W. and J. A. Reinartz. 1988. Sexual dimorphism in biomass allocation and clonal growth of *Xanthoxyllum americanum*. American Journal of Botany 75: 1732-1741.

Queenborough, S. A., D. F. R. P. Burslem, N. C. Garwood and R. Valencia. 2007. Determinants of biased sex ratios and inter-sex costs of reproduction in dioecious tropical forest trees. American Journal of Botany 94: 67-78.

USDA, NRCS. 2009. The PLANTS Database (http://plants.usda.gov, 22 June 2009). National Plant Data Center, Baton Rouge, LA 70874-4490 USA.

Vasiliauskas, S. A. and L. W. Aarssen. 1992. Sex ratio and neighbor effect in monospecific stands of *Juniperus virginiana*. Ecology 73: 622-632.

Vitt, P., K. E. Holsinger and C. S. Jones. 2003. Local differentiation and plasticity in size and sex expression in jack-in-the-pulpit *Arisaema triphyllum* (Araceae). American Journal of Botany 90: 1729-1739.

Warner, R. R. 1988. Sex change and the size-advantage model. Trends in Ecology and Evolution 3: 133-136.

Tools for Assessment of Student Learning Outcomes

1. Pre-lab assessment

A pre-lab assessment is used to prepare students to participate as fully as possible in the field project. Students must read all the instructions in the introduction, along with two background papers on sex change and the size advantage model (Warner 1988; Garcia and Antor 1995), emphasizing that they should focus on the introduction and discussion sections of these two papers. The day before coming to class, students need to fill in a quiz (two short-answer questions) based on these two papers, worth a fraction of the mark of this project and implemented on the course webpage.

These two online questions are: (1) Why is sexual reproduction, both in plants and animals, generally more costly in females than in males (1-3 sentences)? Correct sample answer: Males produce energetically cheap pollen, while females produce comparatively expensive seeds. Female reproduction often entails further costly investment like parental care and provisioning. (2) What does the 'Size-Advantage-Model' propose (1-3 sentences)? Correct sample answer: The size advantage model proposes that when there is a discrepancy between the sexes in the number of expected offspring produced, based on size or age, individuals that can change their sex to take advantage of the discrepancy will maximize their lifetime reproductive success.

2. Data collection and data sharing

Students are assessed for this field project based on the quality of their collected data (large enough sample size, prompt sharing of data). If the field portion of the lab is completed promptly, students can return to the lab and type their results into a spreadsheet to be sent to the instructor/TA. If the field portion of the project runs long and data cannot be entered back in the lab, individual students or each group of students have the responsibility to send a copy of their data in a pre-defined format in an MS Excel spreadsheet by the end of the day to the instructor. This entire data set is then uploaded onto the course webpage for all students to download. I assign a small number of marks for data sharing; if a student or group fails to send data in a timely way, they are penalized for failure to complete this part of the assignment.

3. Written assignment

Students must turn in a written assignment in the form of a short report, not unlike short communications or brief communications in several journals. My intention with this lab report is to encourage students to write succinctly and clearly (they write major reports for other projects in this course). In the short report, students are asked to clearly present: (1) the goals of the study; (2) the methods used; (3) the results, including verbal description of the statistical analyses and the figures through which they test their hypotheses; (4) an interpretation of the results in light of the concept they tested in light of the current literature.

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NOTES TO FACULTY

Challenges to anticipate and solve

1. Challenge #1: ***Finding enough plants.*** Jack-in-the-pulpit is a relatively common plant which typically grows as an understory herb in moist (mixed) deciduous forests of Eastern North America (USDA, 2009). I have run this project in several locations in Southern Ontario and find the species along creeks or otherwise slightly water-logged soils in valley bottoms. Luckily, once suitable ecological conditions and a population of Jack-in-the-pulpit are located, the species tends to be very abundant with enough individuals to (non-destructively and sustainably) service a class of 12 - 16 (or more, if a course is subdivided). Alternatively, sex allocation theory predicts that other perennial dioecious species should also exhibit a similar size distribution between asexuals, males and females and thus another, potentially more abundant local plant species could be used.

2. Challenge #2: ***Search for the inflorescence scar (characteristic of males [but see challenge 5]).*** Instructors should definitely take the class into the field to show students the differences between the three reproductive classes (asexuals, males and females). To spot the male-determining scar just based on illustrations provided in a lab manual is probably not enough and a false scoring of genders should be avoided by any means (but see challenge 5). Once all students have seen the hole and the scar, they experience it as easy to tell the difference between males and females.

3. Challenge #3: Males can have one or two leaves. Males come in different sizes from fairly small to quite large (pers. obs.). Small males tend to have one leaf and then the scar is inserted laterally (Fig. 1) or two leaves where the scar would be inserted between the two leaves, in the same way as for females (Fig.1). Thus students need to be alerted to examine plants carefully.

4. Challenge #4 : Earlier senescence of female leaves. Depending on when the project is done, it might be more or less hard to find females with leaves still attached. Based on my own observations in Toronto (southern-most Canada), female leaves senesce earlier than those of males or non-reproductive individuals. This is probably due to the increased material needs of females for reproduction, i.e. to reallocate resources from leaves into seeds, whereas males and asexuals continue to photosynthesize far into fall or essentially until the first frost. I run this project in the last week

of September and students still find enough females with measureable leaves, even though they often have to assess dead (brown) leaves of females which have dropped to the ground (but still are attached to the infructescence). Because the leaf shape and the leaf venation of Jack-in-the-pulpit are so unique, this early senescence does not pose an insurmountable drawback.

5. Challenge #5: Erroneous scoring of unfertilized females as males. Some female individuals might not receive any pollen and hence not produce any fruits and seeds (or a female might receive only pollen from a close relative, potentially leading to seed abortion due to bi-parental inbreeding). In such cases, female inflorescences, similarly to those of males, would wither and leave behind inflorescence scars which students are requested to look for to determine males. This means that some plants scored as males actually are females. This is an issue which cannot be avoided. However, because the average size difference between the three classes (asexuals, males and females) is so clear, it does not pose a big problem. Nevertheless, the instructor could raise these issues in an open class discussion.

Comments on the Experiment Description

Comments on Introducing the Experiment to Your Students: Because students have been assigned to read two papers, setting the stage and hence initiating preliminary thinking, I generally only briefly ask the class what the framework and the major questions are and how we are going to test them. The instructions for this field project should be fairly straightforward to present to your students. The non-technical nature of the data collection should allow the data collection portion of the lab to proceed relatively quickly and there is, besides the potential erroneous scoring of non-reproductive females as males (see challenge 5), little room for student error.

Comments on the Data Collection: To give the exact description of the data collection sheet to students will help them to structure their data collection and will help you or your TA with the compilation of the class data set. You may want to give the students a blank data sheet appended to this project.

Comments on Questions for Further Thought:

(1) It weakens the difference between males and females and hence the statistical signal, but in most cases this is not a problem. However, students should be aware of this.

(2) It probably will not be able to store away enough resources into its corm that year to use for next year's reproduction and thus probably will turn asexual or male. It might thus take a while before it will be able to put aside enough resources to reproduce as a male or even female again.

(3) Under low light conditions (NB: Jack-in-the-pulpit is fairly shade tolerant) it would probably take much longer for an individual juvenile to become reproductive, i.e. first male and then female. I have had extensions of this basic project where students measured the proportion of open canopy or light conditions above each individual plant, however, there was no significant correlation between the amount of light and reproductive status.

(4) Students need to realize the difference between fixed genders, where a given plant is either a male or a female, and the gender-instability in the case of Jack-in-the-pulpit. Once dioecious (and hence gender-fixed) perennial plants have started flowering, females will, in contrast to males, face this higher cost of reproduction. This results in females investing less than males into above and below-ground organs and their protection, which, in turn, can lead to female-biased mortality (due to hazards of the abiotic and biotic environment). These are thus high follow-up costs.

This issue is "solved" differently in Jack-in-the-pulpit with its labile sex expression. The plant starts out as a small seedling with only limited seed reserves provided by its maternal parent. With this, the seedling can only afford to grow a small leaf, with which it can, in its early life stages and from year to year, only store away a relatively small amount of energy into its corm. This limited amount of energy will hence not allow it to initially start flowering at all, neither as a costly female nor as a less costly male. Only once it can afford to grow ever taller leaves and hence store away some given threshold of energy, the formerly non-reproductive juvenile will switch to being a male. It will continue to express this lower cost gender until it crosses a next threshold to become a female, which can only happen once it grows even larger leaves. Now assume a female suffers herbivory (question 2). With its hence decreased leaf area, it might not be able to store away enough energy to be a female again in the following year. As a reaction, the plant can either turn asexual to amass enough energy for future flowering or it still has enough energy to at least turn male. *The plant hence chooses* *based on its yearly energy reserve the gender it can afford best.* This reasoning is what is encapsulated in the size-advantage model proposed by Warner (1988).

(5) This question points to the heart of frequency-dependent selection. Because only the smallest fraction of individuals of Jack-in-the-pulpit plants in natural populations can afford to be females (i.e. the largest ones), these rare females enjoy the benefits of negative frequency dependence, i.e. that the many males have to fight for access to the fewer females. This in turn means that not all male gametes (pollen) will get access to female gametes (eggs), whereas with an overabundance of males, the probability to be fertilized as a rare egg is relatively high. Hence all individuals of Jack-in-the-pulpit strive to grow tall to become the rare sex. However please note that, despite a general higher frequency of males than females and thus the good chance of being fertilized as a female, Bierzychudek (1981) showed that female plants of Jack-in-the-pulpit tend to be pollen limited, as hand pollinations led to higher seed set.

Comments on the Assessment of Student Learning Outcomes: In the course in which I use this lab unit, I ask my students to write a short report (as opposed to a major report), thus a scientific paper including all normal units of a published scientific publication, but shorter than a major report (1000-1500 words for a short report as opposed to 2500-3000 words for a major report) and with fewer required citations (six in the case of a short report: at least three new ones, whereas a maximum of three can be ‘recycled’ from the lab introduction). Please find instructions on how to write a short report under “1. Instructions for the short report.” I give these instructions also to my students and discuss them in depth, so that they can learn about what characterizes good scientific writing. Following this, I also provide students with a description of how the students’ reports will be graded, under “2. Scoring Rubric.” Such a procedure also provides more transparency in regard to grading and leads to less follow-up questions about marks.

1. Instructions for the short report

1.1. Individual subunits

Title. Concise title potentially containing the main finding of your study.

Abstract. The abstract should explain to the general reader why the research was done and why the results should be viewed as important. It should be able to stand alone; the reader should not have to get any information from the main paper in order to understand the abstract. The abstract should provide a brief summary of the research, including the purpose, methods, results, and major conclusions. Do not include literature citations in the abstract. Avoid long lists of common methods or lengthy explanations of what you set out to accomplish. The primary purpose of an abstract is to allow readers to determine quickly and easily the content and results of a paper. The following breakdown works well: purpose of the study (1-2 sentences), outline of the methods (1-2 sentences), results (1-2 sentences), conclusion (no introduction to this section, no discussion/guesses, no citations).

Key words. List up to 6 key words (fewer key words are OK too). Words from the title of the article may be included in the key words. Each key word should be useful as an entry point for a literature search, pretending your report were to be published.

Introduction. A brief Introduction describing the paper's significance should be intelligible to a general reader. The Introduction should state the reason for doing the research, the nature of the questions or hypotheses under consideration, and essential background. The introduction is the place where you can show the reader how knowledgeable you are with a given field, without being too lengthy. Close the introduction with your main hypothesis/question(s).

Methods. The Methods section should provide sufficient information to allow someone to repeat your work. A clear description of your experimental design, sampling procedures, and statistical procedures is especially important.

Results. Results generally should be stated concisely and <u>without</u> interpretation. Present your data using figures and tables, guide your reader through them.

Discussion. The discussion section should explain the significance of the results. Distinguish factual results from speculation and interpretation. Avoid excessive review. Structure your discussion as follows. 1. First paragraph - restate your major findings concisely, including a statement regarding conclusions you might make regarding your original hypothesis, and then relate to the literature. 2. Discuss the problems that might have been present to influence your findings. 3. Compare your findings with that of others; examine why differences occurred and why this may have been so.

Literature cited. Use the correct format (also see the formatting of the literature in the course manual). You should search for and read related studies beyond those cited below and your report should list at least 6 references, of which 3 should be new (and hence not included in the lab instructions).

1.2. Formatting your report, writing tips

Use the formatting style of the journal "Ecology." It might seem tedious to you to have to follow the many rules the journal prescribes, but adhering to one style makes a paper more organized, increases readability and bad formatting is usually a sign that also the content is of sub-par quality.

Formatting of species names. When mentioning a species in English, also provide the Latin name, at least the first time. Latin names have to be in italics and the first time a Latin name is mentioned, the genus name (first part of the official binary name) has to be spelled out, later on it can be abbreviated, such as in the following example: "Common milkweed, *Asclepias syriaca*, is a hermaphroditic perennial common to Southern Ontario. The leaves of *A. syriaca* are toxic to cattle."

Formatting of references. In the body of the text, references to papers by one or two authors in the text should be in full, e.g. Liang and Stehlik (2009) show *blablabla*. Or: *Blablabla* (Liang and Stehlik 2009). If the number of authors exceeds two, they should always be abbreviated; e.g. Campitelli et al. (2008) show *blablabla*. Or: *Blablabla* (Campitelli et al. 2008). If providing more than one reference in brackets, the order should be chronological with the oldest first and the more recent ones later. In the case of two studies from the same year, the order should be alphabetical. E.g. Blablabla (Zuk 1963; Korpelainen 1998; Stehlik and Barrett 2005, 2006; Stehlik et al. 2008)."

All references cited (and read by you!) in the main text should be included in "Literature cited." References should be in alphabetical order and their formatting should follow the format exemplified below.

<u>Citing articles in scientific journals:</u>

Michaels., D. R., Jr., and V. Smirnov. 1999. Postglacial sea levels on the western Canadian continental shelf: revisiting Cope's rule. Marine Geology 125:1654-1669.

<u>Citing whole books:</u>

Carlson, L. D., and M. Schmidt, eds. 1999. Global climatic change in the new millennium. 2nd ed. Vol. 1. The coming deluge. Oxford Univ. Press, Oxford, U.K.

<u>Citing individual articles/chapters in books (if the individual chapters have different authors than the book):</u>

White, P.S. and S. T. A. Pickett. 1985. Natural disturbance and patch dynamics: An introduction. Pp. 3-13 *in* S. T. A. Pickett and P. S. White, eds. The Ecology of Natural Disturbance and Patch Dynamics. Academic Press, San Diego, California, USA.

Citing a webpage (avoid as much as possible, cite a paper or book instead):

IUCN, Conservation International, and NatureServe. 2004. Global amphibian assessment. Available at www.globalamphibians.org. Accessed October 15, 2004.

Formatting of tables. Tables (if present) should NOT be inserted in your text, but follow, one table per page, after your Literature cited. Give a brief description what the table is about (table caption) and introduce the parameters stated in the table in a text inserted above the table (see examples in all project descriptions). The description should be self-explanatory, thus the reader should not be forced to read the main body of text in order to understand the message of a table. Each column and row in the table should be labeled (with units if necessary). If mentioning a species name, provide the spelled out Latin name (in italics). In the table, round numbers to two meaningful digits.

Formatting of figures. The design of a figure should clearly convey a major result, thus scale your data appropriately. Label all axes with sufficiently large font and meaningful labels. Keep it simple, do not use unnecessary elements such as 3D diagrams if not absolutely necessary based on the data structure.

Similar to tables, figures should NOT be inserted in your text, but follow, one figure per page, after your tables. Give a brief description what the table is about (figure caption) and introduce the parameters stated in the figure in a text inserted below the figure (see examples above). The description of the figure should be self-explanatory, thus the reader should not be forced to read the main body of text in order to understand the message of a figure. Also, each axis in a plot should be labeled (with units) and each bar in a bar chart should be labeled. If mentioning a species name, provide the spelled out Latin name (in italics).

References to tables and figures in the text. In your text, refer to figures as follows: ‘In the spring, temperatures are higher than in the winter (Fig. 1).’ Or: Figure 1 shows that temperatures are higher in the spring than in the winter. In your text, refer to tables as follows: ‘In the spring, temperatures are higher than in the winter (Table 1)’. Or: Table 1 shows that temperatures are higher in the spring than in the winter.

Formatting of statistical references. In the text, the results of a statistical test should be cited in parentheses, in support of a specific statement. Example:

Xylem tension at the top of trees was significantly higher (25 bars) than at the bottom (20 bars) of the tree ($P < 0.05$). When mentioning the result of a statistical test, always provide the P value, R^2 or χ^2 were applicable, mean values, sample sizes and standard errors or confidence intervals. Format your text according to the following example.
"There was a significant difference in the frequency of flowering between low and high elevation sites, with greater bias among low than high elevation populations (average flowering frequency: low elevation = 0.93, SE = 0.01; high elevation = 0.78, SE = 0.02; $\chi^2 = 35.04$, $P < 0.0001$).

Miscellaneous. Avoid quotations - paraphrase your sources instead while making sure you are not plagiarizing.

2. Scoring rubric

1. Information content (30%)

This portion of the grade reflects whether or not you have presented and adequately discussed all of the relevant information. This includes background information on the topic being addressed, as well as the information you have gathered (or should have gathered). Specifically, do not forget to include all relevant statistical result parameters, statistical and other tables, data figures and the written explanation of the results. Also make sure you have cited the adequate number of required articles.

27-30: All of the relevant information was included and discussed adequately.
24-26: One of the pieces of information was not included or discussed adequately.
20-23: One of the most important pieces of information was not included or discussed adequately.
15-20: Two or more of the most important pieces of information were not included or discussed adequately.
<15: Little of the important information was included or discussed.

2. Interpretation and persuasiveness (30%)

This portion of your grade reflects whether or not you interpreted the information correctly and provided persuasive arguments to support your interpretation. Specifically, does your reasoning make sense on its own and also in the light of the published literature, with which you compare your results?

27-30: All of the relevant information was interpreted correctly, and the arguments were very persuasive.

24-26: Most of the information was interpreted correctly, and the arguments were persuasive.
20-23: One of the important pieces of information was not interpreted correctly, or some of the arguments were not persuasive.
15-20: Two or more important pieces of information were not interpreted correctly, and some of the arguments were not persuasive
<15: Little of the information was interpreted correctly, and few of the arguments were persuasive.

3. Clarity of writing (20%)
This portion of the grade reflects whether or not you wrote your sentences and paragraphs clearly. In particular, do you avoid overly long sentences? Are your paragraphs succinct and mostly dealing with one major line of reasoning each? Do your paragraphs preferably start with an introductory sentence and end with a strong summarizing statement? Do you use scientific terms correctly?

19-20: Very clear
16-18: Mostly clear
14-15: Several unclear sentences
10-13: Many unclear sentences
<10: Few clear sentences

4. Formatting (10%)
This portion of the grade reflects whether or not you formatted your report well. This includes the overall structure, the references, and the figures and tables (see instructions below).

9-10: The entire report was formatted correctly, and looked very professional.
8-9: The report was formatted correctly, and looked fairly tidy.
7-8: There were a few formatting errors, or one of the relevant questions was not posed in the introduction.
5-7: There were several formatting errors, or several of the relevant questions were not posed in the introduction.
<5: There were many formatting errors, or few of the relevant questions were posed in the introduction.

5. Spelling, grammar and punctuation (10%)
This portion of the grade reflects whether or not you used correct spelling, grammar and punctuation.

9-10: There were no errors in spelling, grammar, or punctuation
8-9: There were a few minor errors

7-8: There were several minor errors, or a few major errors
5-7: There were several major and minor errors
<5: There were many errors>

Comments on Formative Evaluation of this Experiment:

There are (at least) three specific stages at which students can be given feedback to optimize their learning outcome (points 1 - 3).

1. Design of the data collection sheet
Before students head into the field, they should be taught how to correctly collect data in a spreadsheet in their notebooks. This does not only allow for a more stream-lined assembly of the class data file, but also gives students a basis for better understanding how to go about the statistical analysis. This task could be done in two ways: (1) a minute paper, where each student is individually asked to suggest a layout of the data collection spreadsheet followed by a supervised discussion of the correct format, or (2) by having students work in groups, again followed by a clarifying discussion.

2. Discussion of questions for further thought
The suggested questions for further thought are an excellent opportunity to check whether students have understood what this field project is about and whether they can take what they have learned and develop it further. The instructor could, for example, choose to have students discuss the questions in groups and then merge the several groups into the whole class for a final guided discussion.

3. Feedback on a draft of the short report
In order to maximize the learning effect of the writing assignment, the instructor might consider providing students with feedback on a draft of the paper. This feedback could be provided in several different ways. (1) Through a student peer-review process (minimizing the instructor/TA workload), where, with the help and discussion of the writing tips and the scoring rubric (see **Comments on the Assessment of Student Learning Outcomes**), each student would give feedback to one other student. Using this approach, the instructor should allocate a small fraction of points of the final mark to the peer review. (2) Instructor/TA-based feedback, either in a full-class session, summarizing and discussing common writing problems, or in one-on-one sessions.

Comments on Translating the Activity to Other Institutional Scales or Locations: This lab involves the non-destructive assessment of Jack-in-the-pulpit individuals in forests using a ruler. Multiple lab groups in a larger multisection ecology course could do this lab on separate days or in separate areas of the same forest, as long as there are enough plants.

Some instructors may prefer to lead students to study the hypothesis related to this field project in an inquiry-based approach. In this case, students should be lead to develop the frame-work that males and females face different reproductive costs. They could then be guided to figure out which plant characteristics could be associated with these costs and which could also be measured in the field. Students could consult local plant lists (if available) to identify local dioecious species and search for them in the field. Students then should develop testable hypotheses associated with the identified study species and plant characteristics. They would then collect and analyze data. The instructor would discuss with students their findings along the way and could guide students to appropriate citations in the literature concerning costs of reproduction.

This lab could be easily used in upper level high-school settings. The statistical analysis could be merely descriptive by plotting mean leaf sizes per reproductive group.

GENERIC DISCLAIMER

Adult supervision is recommended when performing this lab activity. We also recommend that common sense and proper safety precautions be followed by all participants. No responsibility is implied or taken by the contributing author, the editors of this Volume, nor anyone associated with maintaining the TIEE web site, nor by their academic employers, nor by the Ecological Society of America for anyone who sustains injuries as a result of using the materials or ideas, or performing the procedures put forth at the TIEE web site, or in any printed materials that derive therefrom.