Samara Dispersal in Boxelder: an Exercise in Hypothesis Testing

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Traditional approaches to teaching botany, with their emphasis on preserved specimens, prepared slides, and slow-to-develop physiological experiments tend to lead students to one conclusion—plants are boring (Uno, 1994). Surveys have also shown that students have a strong preference for hands-on experiments which utilize living material and generate fast results. The incorporation of such experiments into botany or ecology exercises is especially difficult in the "dead" of winter in northern climes. We present here a fun, inexpensive, and pedagogically useful laboratory exercise which involves indoor studies of the dispersal properties of the winged fruits (samaras) of boxelder (*Acer negundo* L.) trees. The exercise engages students in the processes of hypothesis testing, experimental design, and data analysis, as well as introducing them to some important concepts relating to functional ecology, succession, and plant reproductive biology.

Boxelder Samaras

Samaras are winged fruits morphologically adapted for wind dispersal because they autorotate when falling. The functional significance of autorotation is to increase the distance the fruit may be transported by horizontal winds (Augspurger, 1986). The longer a samara is airborne, the greater are its chances for settling in a gap in the forest canopy far from the shading limbs of the maternal plant.

A large and variable percentage (20 to 60% in our experience) of boxelder samaras are

sterile. Isolated specimens tend to have a higher percentage of sterile samaras. Unlike most plants, boxelders retain their sterile fruits rather than aborting them, and these sterile samaras develop in parallel with the fertile samaras, and their appearance is indistinguishable.

Boxelders are commonly found growing along riverbanks, or along the borders of moist fields or railroad embankments. Boxelder often serves as an invading pioneer species during the succession of old fields. It is advisable to collect the samaras in the early autumn, and let them dry uniformly before use.

Three Hypotheses

In this exercise, the students are asked to consider three different hypotheses concerning the respective dispersal of sterile and fertile boxelder samaras.

Hypothesis I is based on the assumption that it would be advantageous to have sterile fruits fall closer to the maternal plant. Because sterile fruits have zero reproductive potential, they are essentially a lost investment for the maternal plant. Some of this lost investment might conceivably be re-cooped if the sterile fruits contributed to the "leaf-litter" immediately beneath the maternal plants. Indeed, Otto & Nilsson (1981), in attempting to explain the winter-time retention of lower canopy leaves in beeches and oaks, proposed that the delayed shedding of the previous year's leaves may benefit the tree by providing the nearby ground, well-leached by winter's thaw, a fresh supply of soluble nutrients in the early spring. *Hypothesis I, therefore, predicts that the optimal reproductive strategy for species such as boxelder, would be to have fertile samaras land farther from the parental plants, and to have sterile samaras fall closer.*

Hypothesis II is based on the notion that ability of boxelder seeds to colonize an old field is limited chiefly by post-dispersal seed predation. Studies of old field colonization have shown that the largest mortality of colonizing tree species is due to predation of seeds by rodents (Gill & Marks, 1991). Conceivably, the prolonged dispersal of boxelder fruits over the course of winter may be an evolutionary adaptation for reducing seed predation by winter-foraging mice. The "cryptic sterility" of boxelder samaras may also lower their desirability as a food source and may act, therefore, as an effective adaptation for lowering post-dispersal seed predation. If so, then this strategy of "cryptic sterility" would be most effective if the post-dispersal mixing of sterile and fertile samaras were maximal. *Hypothesis II, therefore, predicts that sterile seedsshould fall the same distance from the parent as do fertile samaras*.

Hypothesis III is based on the findings of researchers such as Green (1980) who have found that the rate of descent of samaras is highly correlated with the square root of the samara's wing-loading (samara mass divided by wing-surface area). For example, Augspurger and Hogan (1983) found that fruits of *Lonchocarpus* with 0, 1, 2, or 3 seeds had increasingly large values both for square root of wing loading and for rate of descent in still air. Therefore, based on the reasonable assumption that fertile samaras weigh more than sterile ones, *Hypothesis III predicts that fertile samaras, bearing more wing load, should fall closer to the parent than do sterile samaras.*

After having the three hypotheses explained to them, the students are asked to break into groups of three and devise an experiment to decide between the three hypotheses.

Experimental Design

Based on their training and inclinations, the students propose a multitude of diverse approaches, some more feasible, and some more imaginative than others, to test the predictions of the three hypotheses.

One of the more common suggestions involves collecting the fallen fruits in transects surrounding specimens of boxelders, noting the distances between the site of collection and the presumed parent plants, and then determining the reproductive status of the fruit. Although this research strategy has the advantage of being most natural, questions arise over the uncertainty of assuming that the closest specimen is indeed the mother of all the collected fruits and, whether the presumably more nutritious fertile fruits might not be subject to enhanced predation. Although both of these problems are surmountable, their resolution might require many months of outdoor study. It is more feasible to study the question indoors using collected fruits.

Another common proposal involves measuring the differences in the dispersal properties of fertile samaras before and after the seeds have been surgically removed. This approach, too, is discouraged because it is no trivial matter to remove the seed without damaging the samara and, moreover, such manipulations might well alter the samaras' aerodynamic properties.

Students must also confront the problem of what parameter to measure. Many students propose measuring the horizontal distance a dropped samara travels from the plumbline. However, because of the vagaries and weakness of indoor air currents, this approach, too, is discouraged: The samaras when dropped in still air, generally fall close to the plumbline. The simplest and most straightforward suggestion is to measure the time needed for the descent of dropped samaras in still air, and then to determine whether they are sterile or fertile.

Students are also confronted with problems associated with the statistical analysis of their data. One of the more common sources of error in data collection and analysis arises from the non-independence of data values (*i.e.*, pseudoreplication). Students are often tempted to treat repeated measurements taken from the samara as independent values during statistical analysis. This error arises from the misconception that the aim of scientific observation or experiment is to obtain large number of measurements rather than measurements from a large number of subjects. True replication of measurements, however, is necessary to guard against aberrant single values. We recommend that each group of students measure the descent time of 30 different samaras three times each, and determine the average descent time for each of the 30 samaras.

We have found that students can get useful results by standing atop laboratory benches (much to their delight) and dropping samaras from a position parallel and adjacent to the ceiling. (For safety concerns, the samara droppers should wear suitable shoes while standing atop the table). A second student measures the time of descent with a stopwatch (0.01 second resolution), while a third records the data. To avoid mistakes and confusion, it is a good idea to have the students number their samaras with a fine-tipped, indelible marker prior to experimentation. Students should be reminded that each samara should be dropped and timed in a uniform manner to reduce experimental error. The exercise can also be expedited by supplying the students with

a standard data sheet. The significance of the difference between the fertile and sterile samaras can be analyzed by a two-sample t-test (see Table 1).

Another salient property that can be measured is samara mass (Table 1). Usable measurements can be achieved with a typical laboratory pan balance with a resolution of 0.01 g. Not too surprisingly, fertile samaras weigh more on average than do sterile samaras (Table 1). Plotting samara mass v. time needed to descend reveals a clear inverse relationship (r = -0.66) between the two: fertile samaras fall faster (see Fig. 1). The predictions of Hypotheses I and II are rejected, and the prediction of Hypothesis III is supported.

In closing we refer readers to an earlier How-to-Do-It publication (Thomson & Neal, 1989) which presents additional pedagogical exercises pertaining to the wind dispersal of fruits and seeds, including the construction of artificial paper samaras, studies of the effects of partial dissections on the dispersal properties of tree-of-heaven (*Ailanthus*) samaras, and the collection in transects of spraypainted samaras dropped from a flagpole.

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	N	Mass	Area	Descent
		(gm)	(cm^2)	Time (sec)
Fertile	72	0.073 ±	3.27 ±	3.11 ± 0.25
	(200)	0.007	0.35	
Sterile	28	0.051 ±	3.24 ±	3.65 ± 0.23
an a secondar daar - daar maaring		0.006	0.42	
	0			
T-Test		T=15.23** p<0.001	T=0.89 p>0.05	T=19.29** p<0.001

Table 1: Summary of experimental results demonstrating that fertile boxelder samaras on average weighed more and descended a distance of 2.4 in more quickly than did sterile samaras. No significance difference was found between the average surface areas of fertile and sterile samaras.



Figure 1: Relationship between boxelder samara mass and time needed to descend 2.4 m (n=100). Note the clear separation between fertile and sterile samaras.