

GSAS Merit Fellowship: Statement of Purpose

Daniel Buonaiuto, *Organismic and Evolutionary Biology*

Introduction

Models of growth and development shape our understanding of plant ecology and evolution, and determine projections of plant biology with future climate change. In a typical model of plant development, vegetative growth precedes reproduction. However, for as many as 30% of trees and shrubs of eastern North America, it is not the green tips of leaves that mark the start of the growing season, but the colors of flowers (Buonaiuto *et al.*, 2020). Why, and how, do so many species in these regions produce their flowers before their leaves; during the time of year when risk of frost damage is the highest (Augspurger, 2013) and their energy reserves are at their lowest (Primack, 1987)? How will climate change alter these patterns, and what are the implications of these changes for plant fitness and ecosystem functioning in the coming decades?

My PhD is focused on answering these fundamental and applied questions about the flower-leaf sequences (FLSs) of deciduous woody plants. Funding from the *Graduate School of Arts and Sciences* would allow me to undertake a project that will address these questions at an unprecedented scale and fill in several important gaps in our knowledge about this unique plant trait.

Background

Several hypotheses have been put forth in the literature to explain this risky behavior of flowering prior to leaf growth. Flowering during the leafless season may allow for efficient pollen transfer in wind-pollinated species (Rathcke & Lacey, 1985) or reduce drought stress by partitioning the evaporative demand of flowers and leaves across the seasons (Gougherty & Gougherty, 2018). Flowering-first may also simply be a byproduct of extremely early flowering (Primack, 1987) and variation in FLS patterns among species could be more strongly determined by developmental or evolutionary constraints than ecological function (Diggle, 1995; Gougherty & Gougherty, 2018).

In a previous project I used statistical modeling to compare and evaluate these hypotheses (Buonaiuto *et al.*, 2020). Across all of my models, I consistently found a strong association between wind-pollination and the flowering-first FLS. I also showed that early flowering and phylogeny, the shared evolutionary lineages among taxa, strongly influence this trait. This support for multiple hypotheses suggests that flowering-first FLSs may have evolved multiple times under different environmental conditions and raises new questions about the functional significance of this trait.

In particular, the strong support for the wind-pollination hypothesis from my models offers little information about the functional significance of flowering-first in the many insect-pollinated species share this FLS. To fill in this gap, I intend to undertake a new project that will explore the functional ecology of flowering-first FLSs in insect-pollinated taxa by exploring associations between FLSs, species distributions and functional traits using a large data set of herbaria specimens. I present the details of this project below.

Project Proposal: The functional ecology of flower-leaf sequences in the American plums

The American plums (*Prunus* subsp. *Prunus* sect. *Prunocerasus*) are an ideal group to use to address my research question. The section consists of 16 species that are distributed across North America and vary in their FLSs (Shaw & Small, 2004). The Harvard herbaria houses 3,134 digitized specimens of these species and the Consortium of Midwest Herbaria collection contains an additional 13,869. Leveraging these 17000+ specimens, I will test the major hypotheses of the function of FLS variation in insect-pollinated taxa by testing for correlations between FLS patterns and the functional traits and ecological requirements of these species.

Part I: Quantification of FLS variation

A major challenge to study FLS variation is that most data sources describe FLS patterns using broad verbal descriptions (e.g. “flowers before leaves”). These categories are imprecise and, unsurprisingly, different sources classify the same species differently (Buonaiuto *et al.*, 2020). While its possible that these differences could reflect temporal or geographic variability in FLSs, they could equally be the product of observer bias. It is important to apply a more rigorous quantitative method for describing FLSs.

To characterize FLS patterns among *Prunocerasus* species, I will score the vegetative and flowering stages of 500-1000 herbarium specimens per species using the BBCH scale (Finn *et al.*, 2007), a standardized method for robustly evaluating the timing of seasonal events in plants. From these data, I will use Bayesian generalized linear models to predict a likelihood that a given species flowers before its leaves emerge, providing the first quantitative estimates of FLS variation for these species, and demonstrating a robust method to do so for other taxa. Using this method, I will also be able to quantify variation in FLS patterns within species and test for any temporal or environmental covariates that may drive this variation.

Part II: Assessing FLS hypotheses

Each of the dominant hypotheses of the functional ecology of FLS variation makes a prediction about the environmental characteristics or biological traits that should be associated with flowering-first species.

The **water limitation hypothesis** (Gougherty & Gougherty, 2018) predicts that the geographic ranges of leafing-first species will be more limited by water availability than flowering-first species. To test this hypothesis, I have obtained measurements of the historic drought conditions (based on the Palmer Drought Severity Index (Dai *et al.*, 2004)) at the collection site of each

herbarium specimen and will test for differences in aridity tolerance across geographic ranges of predominantly flowering-first and leafing-first taxa.

The **early flowering hypothesis** suggests that flowering-first is an adaptation that allows for extended fruit development time (Primack, 1987), and predicts that flowering-first species will have larger fruits than leafing-first species. To test this hypothesis, I will measure the diameter of fruits present on herbarium specimens and test for differences between predominantly flowering-first and leafing-first taxa.

The **insect-visibility hypothesis** suggest that flowering-first species are easier for insect pollinators to locate (Janzen, 1967) and predicts that flower displays will differ in size between flowering-first and leafing-first species. To test this hypothesis, I will measure the corolla diameter of 5-10 flowers from 50-100 herbarium specimens/species and compare the flower size between predominantly flowering-first and leafing-first taxa.

Scope and implications of the project

I have tested the methods described above in a pilot study of a subset of 200 *Prunocersus* herbarium specimens and found the methods are robust for both quantifying FLS patterns among species and testing the FLS hypotheses through trait associations. Expanding this effort to evaluate 8,000-16,000 specimens is an ambitious goal, but my pilot study suggest that I could complete this project in a semester with the support of the GSAS merit fellowship allowing me to make this work my primary focus.

This project will fill in a major gap in the basic understanding of the functional significance of flower-leaf sequence variation in deciduous woody plants. It will unite the underlying physiology and ecology that drives FLS variation, and provide a path forward for predicting the scope and implications of FLS shifts with climate change.

References

- Augspurger, C.K. (2013) Reconstructing patterns of temperature, phenology, and frost damage over 124 years: Spring damage risk is increasing. *Ecology* **94**, 41–50.
- Augspurger, C.K. & Zaya, D.N. (2020) Concordance of long-term shifts with climate warming varies among phenological events and herbaceous species. *Ecological Monographs* **n/a**, e01421.
- Buonaiuto, D.M., Morales-Castilla, I. & Wolkovich, E.M. (2020) Reconciling competing hypotheses regarding flower–leaf sequences in temperate forests for fundamental and global change biology. *New Phytologist* **n/a**.
- Dai, A., Trenberth, K.E. & Qian, T. (2004) A global dataset of palmer drought severity index for 1870–2002: Relationship with soil moisture and effects of surface warming. *Journal of Hydrometeorology* **5**, 1117–1130.
- Diggle, P.K. (1995) Architectural effects and the interpretation of patterns of fruit and seed development **26**, 531–552.
- Finn, G.A., Straszewski, A.E. & Peterson, V. (2007) A general growth stage key for describing trees and woody plants. *Annals of Applied Biology* **151**, 127–131.
- Gougherty, A.V. & Gougherty, S.W. (2018) Sequence of flower and leaf emergence in deciduous trees is linked to ecological traits, phylogenetics, and climate. *New Phytologist* **220**, 121–131.
- Janzen, D.H. (1967) Synchronization of sexual reproduction of trees within the dry season in central america. *Evolution* **21**, 620–637.
- Primack, R.B. (1987) Relationships among flowers, fruits, and seeds. *Annual Review of Ecology and Systematics* **18**, 409–430.
- Rathcke, B. & Lacey, E.P. (1985) Phenological patterns of terrestrial plants. *Annual Review of Ecology and Systematics* **16**, 179–214.

Shaw, J. & Small, R.L. (2004) Addressing the "hardest puzzle in american pomology:" phylogeny of prunus sect. prunocerasus (rosaceae) based on seven noncoding chloroplast dna regions. *Am J Bot* **91**, 985–996.