Proposal for the Research, Scholarship and Creativity Grant. 2008-2009.
Dr. Pamela M. Kittelson, Department of Biology, Gustavus Adolphus College.

A. Purpose:

I intend to explore how long-term temperature trends may alter flowering dates, and how variation in plant life histories and physiological traits influence flowering phenology. The objective of this project is to analyze a long term data set (15 year) with my collaborator at the University of Montana to determine if flowering times for 35 herbaceous perennial species have changed over time and are correlated with changes in temperature. Additionally, we are interested in determining if physiological and plant life history traits may explain patterns in flowering phenology. Little research has attempted to characterize flowering time for a group of co-occurring species. This proposed collaboration will enable me to conduct research related to ecological physiology and plant monitoring, which will inform my teaching practices.

B. Feasibility:

Qualifications

- 15 years of research experience related to plant physiological ecology.
- Ph.D. in botany and ecology.
- A funding and publication record that demonstrates an ability to transform proposed hypotheses into completed experiments, and ultimately into peer reviewed publications. All former R.S.C.s have resulted in published, peer-reviewed manuscripts.
- A demonstrated ability to work collaboratively; three of my recent published manuscripts were collaborations done with researchers from the University of Montana.
- Three summers of other research in the montane grasslands of western Montana.
- A demonstrated ability to incorporate techniques and literature into courses I teach.

Preparation:

I have been invited to collaborate with Peter Lesica at the University of Montana where I was a visiting researcher during my last sabbatical. Peter is a well-respected Montana botanist with >80 peer-reviewed publications about Montana flora and ecology. For the past 15 years he has kept records of flowering time from surveyed transects. Peter would like to collaborate with a plant physiological ecologist (me) in order to analyze the data with an eye toward addressing if plant life history traits may explain plasticity in flowering dates for some species.

Time period:

June 2008- May 2009. We have begun entering our demographic data into spreadsheets, but I do not have time to analyze these data until summer. I will travel to Montana in order to collect July/August data, to use the University library and its deep databases, to take physiological measurements of the plants in the field, to analyze data, write and revise drafts with my colleague. Please refer to the schedule in the project design section. My colleague and I fully anticipate that this time period is sufficient to complete the analyses and writing.

C. Project design:

An organism's life history characteristics are comprised of a suite of demographic traits related to reproduction and survivorship that have evolved in response to environmental factors (Roff

1992). Life history strategies are an assemblage of traits that represent trade-offs in energy allocation for a given habitat (Stearns 1992). For example, in fluctuating environments species might trade-off early flowering and high reproductive output with lower adult survivorship; in contrast, individuals that mature later in life tend to produce fewer offspring, but they tend live longer. These life history strategies emerge through many generations of selective pressure. Thus, life history strategies are shaped by variation in the environment.

One major life history trait is flowering development and timing, which is termed flowering phenology. For some perennial species, flowering time can be triggered by genetically predetermined switches that are independent of environmental cues (Woodward et al 1997). In other species, flowering switches also can be turned on only by a specific daylength or the number of days a plant has been dormant (Hopkins 2004). For still other species, flowering time is very plastic and a function of fluctuating environmental cues; warm soil temperatures and high levels of precipitation can alter the date that a plant flowers in any given year (Stanton et al. 2000). Finally, flowering date can be triggered by a combination of genetically entrained switches and environmental cues. Thus, perennial plant species sit on a continuum that ranges from genetically-fixed flowering dates to purely environmentally-determined flowering dates. Few researchers have collected long-term data sets to compare and contrast flowering time patterns in one plant community or used these data to correlate trends in flowering to environmental variables such as temperatures, and then attempted to link these patterns to life history strategies and physiological ecology.

Intermontane grasslands of western Montana have experienced a mean average temperature increase of 0.9 degrees C over the past 20 years (relative to 180 year average; Martin 2007, Prentice et al. 1991). Spring is coming earlier to western Montana grasslands, and the annual summer drought is also occurring about 10-15 days earlier. Plants that have a flowering time which are fixed and determined only by daylength will not flower earlier despite changing environmental conditions and these species could risk reductions in reproductive fitness (flowering too late could mean that many plants will not have adequate water resources to complete reproduction or seed production will be reduced). In contrast, plants that possess a flowering time that is determined in part by environmental cues could potentially adjust to earlier springs and summer drought, ultimately resulting in a higher relative fitness. Over time, plant species that are more phonologically plastic may produce more seeds and could become competitive dominants, thereby shifting plant community composition. Understanding which plants can alter flowering phenology relative to changing environmental conditions can provide us with a capacity to predict changes in community composition as the climate continues to warm. Moreover, generalities among the ecophysiological and life history traits may provide us with an ability to predict patterns for other plants in other communities.

This experiment specifically addresses the following questions related to flowering phenology, plant life history and physiological traits of 35 montane grassland species.

1) What the date of first flower for each plant species? Has the date changed over time?

2) For each species, does the date of first flower correlate with average temperature trends or warming degree days? For example, in warmer springs do some species tend to flower earlier? Which species appear to have fixed flowering phenologies independent of temperature trends?

3) What life history traits correlate with flowering time 'behavior'? Are species with a flexible flowering time smaller, more fecund and shorter-lived than individuals with a fixed phenological trend (as life history theory would predict)?

4) What physiological traits correlate with flowering time 'behavior'? Physiological traits include: carbon to nitrogen ratios, root: shoot ratio, water use efficiency and photosynthetic capacity.

The results of these studies will help connect physiological processes and life history traits with trends in flowering time. Flowering phenologies for 35 species will be characterized and the relative flexibility of the phenology relative to temperatures will be better understood. These results may help us identify underlying, mechanistic processes that ultimately influence whole plant ecological response. Additionally, predictions can be made about potential community-wide responses to sustained and higher spring temperatures.

<u>Schedule of activities at University of Montana –library, herbarium and our field site nearby</u>

- Measure physiological and life history traits in the field. July-August 2008.
- Collect flowering time data for July-August 2008.
- Analyze data from the past 15 years; incorporate information on physiological and plant life history traits. July 2008-May 2009. In Montana and at Gustavus.
- Write and edit. August 2008-May 2009. In Montana and at Gustavus.
- Use library and databases to research information on the 35+ species. July - August 2008.

<u>Anticipated outcomes</u>

- An article for publication in a peer reviewed journal, e.g. American Journal of Botany.
- Continue practicing techniques and statistics related to plant physiological ecology.
- Investigate new literature in plant ecophysiology some of which can be used in directed discussions in plant physiology (Bio378) and ecology (Bio370).

D. Literature cited

Hopkins, H. 2004. *Plant Physiology*. Prentice-Hall.

Martin, T. E. 2007. Climate correlates of 20 years of trophic changes in a high-elevation riparian system. Ecology 88: 367-380.

Prentice, I. C., P. J. Bartlein and T. W. Webb III. 1991. Vegetation and climate change in eastern North America since the last glacial maximum. Ecology 72:2038–2056.

Roff, D. A. 1992. The evolution of life histories. Chapman and Hall, New York.

Stanton, M. L., B. A. Roy and D. A. Thiede. 2000. Evolution in stressful environments. I. phenotypic variability, phenotypic selection, and response to selection in five distinct environmental stresses. Evolution 54: 93-111.

Stearns, S. C. 1992. The evolution of life histories. Oxford Univ. Press, Oxford, U.K.

Woodward, F. I. and C. K. Kelly. 1997. Plant functional types: towards a definition by environmental constraints. Pp. 47–65 *in* T. M. Smith, H. H. Shugart, and F. I. Woodward, eds. *Plant functional types: their relevance to ecosystem properties and global change.* Cambridge Univ. Press, Cambridge, U.K.

PREVIOUS PROJECT SUPPORT

I received Research, Scholarship and Creativity Grants in 2000, 2002 and 2005.

2000 RSC GRANT: My students and I completed all our proposed research. We established permanent sampling plots to monitor flowering plant diversity in mowed and burned prairies at three prairie restoration sites in southern Minnesota. Results from these experiments clarified the effects of fire and simulated grazing on forb diversity. Our project was designed to help define the best course of action for the Linnaeus Arboretum and long-term data were beneficial to Department of Natural Resources and The Nature Conservancy.

Outcomes achieved from 2000 R.S.C.

- Established 90 permanent plots; burned and mowed.
- Censused above-ground diversity; measured plant biomass and N content
- Censused seed bank, seedling density and survivorship
- Provided management plan to Arboretum prairie that was implemented.
- Presented findings at multiple symposia: Sigma Xi (spring 2001 and 2002), NCUR (spring 2002) and Ecological Society of America (summer 2002).
- RSC-funded results led to continuation of faculty/student research funded through the Rockefeller Foundation and administered by Environmental Studies ($16,000).
- Student assistants supported through RSC 2000 grant: Nicole Barondeau, Leslie Brandt, Jim Eckberg, Phil Graeve, Amber Krahmer, Ginger Lindgren, and Monica Paulson.
- An article from this work focusing on use of indicator species is currently in a second round of revision for the Journal of the Iowa Academy of Sciences
- J Eckberg, E. Bockman and I published, "Adding nitrogen controls invasive yellow sweetclover, *Melilotus officinalis*," in Ecological Restoration (25: 279-281).

2002 RSC GRANT: I completed everything that I proposed in the grant. My proposed research was focused on three outcomes: 1) learning and practicing a new molecular technique for population analyses called AFLP (Amplified Fragment Length Polymorphisms); 2) identifying AFLP markers to use for population genetic analyses, and; 3) incorporating these new techniques into my teaching, research and ultimately, grant proposals.

Outcomes achieved from 2002 R.S.C.

- I attended a three-day intensive AFLP workshop offered by Biotechnologies for Ecological, Evolutionary, and Conservation Sciences at the University of Florida.
- Collected leaf and seed tissue from three populations; extracted DNA (n=300 samples)
- Traveled to Moss Landing Marine Labs in California; practiced the AFLP technique.
- Optimized 'recipes' to carry out the analysis.
- Investigated and applied for funding. Colleen Jacks, Jeff Dahlseid and I were awarded a LiCor automated DNA sequencer ($60,000).
- Incorporated concepts into 200-300 level biology courses (especially Bio 396).
- Taught the technique to seven undergraduate research students; student posters at Ecological Society of America (Stephen Handler; Summer 2004) and on-campus symposium (Josh Dwyer, Angela Ingersoll, Elaine Mans and Jenn Rieke; Fall 2004).
- In 2006 Stephen Handler and I had an article accepted for publication titled "Genetic diversity in isolated patches of the tallgrass prairie forb, *Lithospermum canescens* in the Journal of the Torrey Botanical Society (133: 513-518).

2005 RSC GRANT: During the academic year 2005/06 I spent my sabbatical at the University of Montana, where I conducted collaborative research with John Maron, Marilyn Marler and Dan Mummey. The RSC supplemented a NSF Research Opportunity Award and helped me set-up and maintain an experiment with three primary objectives: 1) to compare the impact of three invasive species on survival, biomass and leaf construction of four native species established assemblages that differed in native species diversity, 2) to examine how native and invasive species identity influenced mycorrhizal diversity, and how invaders altered mycorrhizal diversity, and 3) to incorporate knowledge gained from this research into my teaching and to further my current research trajectory. Our results identified underlying, mechanistic processes that ultimately influence whole plant ecological response to invasive species.

<u>Outcomes achieved from 2005 R.S.C.</u>

- Completed all data collection for experiments in the lab, greenhouse and field.
- Learned how to determine water and nitrogen use efficiency.
- Re-familiarized myself with equipment that measures photosynthetic capacity and water potential.
- Gave a Shoptalk about results from one of these experiments.
- Developed and held a community outreach activity that discussed the importance of mycorrhizal interactions with plants. Missoula County Extension and Prairie Keepers.
- Will give a seminar at the Northwest Scientific Association meetings in March 2008
- Provided a research experience for one student (Chris Pinahs '06) in January 2006.
- Developed two new ecological physiology labs titled: 'Physiological and morphological plasticity within populations of dandelions' and 'Water use efficiency' for use in Plant Physiology (Bio 378), and two new case studies called 'Effects of invasive species' and 'Are some communities more invasible than others?" for Ecology (Bio 370) and Biodiversity (Bio100).
- Investigated new literature in plant ecology and ecophysiology for use in directed discussions in plant physiology (Bio378), EEB (Bio202) and ecology (Bio370).
- Was the primary author of a manuscript that was accepted in Ecology (in press for May 2008) titled "An invader differentially affects leaf physiology of two natives across a gradient in diversity." Second and third authors: John Maron and Marilyn Marler.