

Overview of experimental setup, each pot contains one needle goldfield population and one biodiversity treatment.

**STUDENT RESEARCH**  
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**EDITH LAI**

# Biodiversity Loss and Phenology in California Grasslands

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## Background

A major consequence of climate change on plant communities is its effect on phenology - the seasonal timing of life events such as germination, flowering, and fruiting. The main factors influencing plant phenology are temperature, photoperiod, and precipitation, but the effects can be compounded by additional factors (Parmesan and Yohe 2003, Franks 2011). Studies have concluded that the general phenological response to global warming is a shift toward earlier flowering seasons as a result of warmer temperatures during winter (Parmesan and Yohe 2003, Cleland *et al.* 2006 and 2007, Menzel *et al.* 2006). This shift is significant because seasonal synchronicity between plants and pollinators or between plants and consumers may be disrupted, potentially causing complex and adverse bottom-up effects in higher trophic levels (Franks 2015). A recent study demonstrated that declining biodiversity also affects flowering in California grasslands through direct manipulation of species composition in grassland reserve plots over the course of four years. Multiple species flowered significantly earlier in response to reductions in diversity, suggesting a stronger role of biotic factors in phenology (Wolf,

Zavaleta, and Selmants 2017). The advance in flowering caused by biodiversity loss is comparable to the advance caused by changing physical conditions attributed to global warming. In addition, biodiversity is positively associated with ecosystem productivity and stability over extended periods of time (Tilman, Reich, and Knops 2006, Craven *et al.* 2018). With downstream effects of climate change leading to invasive species introduction and habitat destruction, declining biodiversity is a significant threat to many ecosystems (Butchart, Walpole, and Collen n.d., Dukes 2002). The combined effects of increasing temperature, precipitation change, and biodiversity loss will have severe effects on California grasslands; therefore, further study on the relationship between biodiversity and phenology is integral to informing the land management and conservation of this valuable ecosystem.

This study is framed around needle goldfields (*Lasthenia gracilis*), an annual herb found throughout California grasslands. This species was chosen because of its widespread distribution and environmental tolerance. In addition, high trait variation and high fixation index — the genetic distance between populations — indicate significant population level differences (Montalvo, Riordan, and Beyers 2017). Within the *Lasthenia* genus and multiple other genera, populations found in decreasing latitudes correlate with earlier flowering time (Hall *et al.* 2007, Olsson and Ågren 2002, Olliff Yang unpublished data). In regions closer to the equator, limited water availability due to dry summers prompts populations to flower earlier, a response called

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A monoculture pot of Lost Hills population flower.

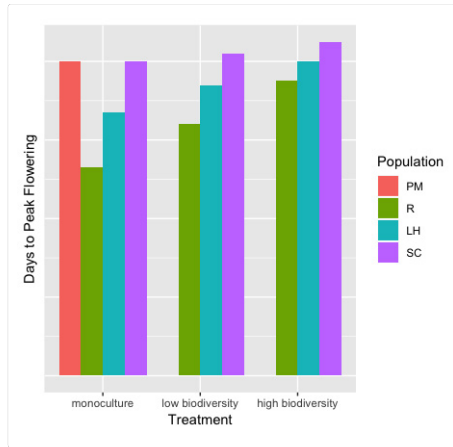


Figure 1. Hypothesized trends in peak flowering date in relation to biodiversity.



Figure 2. Latitudes of needle goldfield populations included in study.

## Biodiversity Loss and Phenology in California Grasslands *continued*

drought escape (Franks 2011, Shavrukov *et al.* 2017). In Central Valley and coastal regions of California, where grasslands are located, average annual precipitation decreases as latitude decreases (California Department of Water Resources). Lower latitude plants benefit from early flowering by being able to complete their life cycles early while water is still plentiful. A study on phenotypic plasticity found that among sagebrush, lower latitude populations display greater plasticity in flowering time than higher latitude populations in response to photoperiod and temperature (Richardson *et al.* 2017). Flowering time plasticity, which is the variance in the timing of flowering expressed in response to differential conditions, can be a beneficial adaptation in changing environments. This experimental study seeks to determine if a relationship exists between the latitudinal gradient of populations within a single species and the phenological response to biodiversity loss. Given that lower latitude plants have adapted to flowering earlier to maximize water availability and the phenological plasticity involved in drought escape response, lower latitude populations may exhibit greater phenological plasticity in response to declining biodiversity. To test this, populations of needle goldfields chosen from three latitudes of California grasslands were grown in environments with decreasing levels of biodiversity. We hypothesized that declining biodiversity would correlate with greater phenological shifts for needle goldfield populations originally from lower latitudes than needle goldfield populations originally from higher latitudes in California. The expected results are displayed in Figure 1, with R, LH, and SC representing populations of increasing latitude and PM representing a monoculture mixture of the three populations. In addition, we hypothesized that flowering season lengths for populations would decrease in response to declining biodiversity due to the positive relationship between biodiversity and ecosystem productivity.

### Methods

#### Setup

This experiment took place within a lath house at the University of California, Berkeley, Oxford Tract facilities. Lath houses allow for the

use of natural climate and precipitation while protecting the experiment from extreme disturbance. Needle goldfield seeds provided by Pacific Coast Seed originated from Santa Clara, Lost Hills, and Riverside counties to represent populations of high, medium, and low latitudes, respectively (Fig. 2).

The other species used to represent a California grassland community were annual lupine (*Lupinus bicolor*), small fescue (*Festuca microstachys*), dotseed plantain (*Plantago erecta*), and annual hairgrass (*Deschampsia danthonioides*). These species were chosen from a larger pool of common grasses and annual herbs. Each population of needle goldfields received three biodiversity treatments: monoculture, low biodiversity, and high biodiversity in a fully crossed design. The masses of seeds in each pot were standardized to control for density dependent effects. In monoculture treatment, 170 mg of needle goldfield seeds were planted in a single pot. In low biodiversity treatment, 56.7 mg each of needle goldfields, annual lupine, and small fescue seeds were planted together in the same pot. In high biodiversity treatment, 34 mg each of seeds of all five species were planted in the same pot. An additional treatment group of mixed populations contained equal amounts of needle goldfield seeds from each of the three populations to compare the phenological effects of population level genetic diversity with community level species biodiversity. Each population and treatment were grown in six-inch diameter pots with high drainage soil. There were five replications of each population and treatment, totalling to 50 pots of five mixed monoculture populations and 15 pots of each needle goldfield population and biodiversity treatment.

#### Data Collection

Seeds were planted in pots in early February and regularly watered throughout the spring season. After observing the first emergence, we counted the number of emerged needle goldfield seedlings in each pot biweekly to determine the peak emergence dates for each treatment.

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# Biodiversity Loss and Phenology in California Grasslands *continued*

After emergence, we collected data on needle goldfield flowering by counting the number of buds and open inflorescences. Data collection concluded in late June, following the conclusion of the typical needle goldfield bloom period and the senescence of all plants.

Phenology measurements included peak emergence date, first flowering date, peak flowering date, and flowering season length. First flowering is the date in which the first flower opened within a pot, while peak flowering date and flowering season length were determined by counting active reproductive structures — both buds and inflorescences. The flowering season was defined as the number of days between the first and last flower openings.

## Analysis

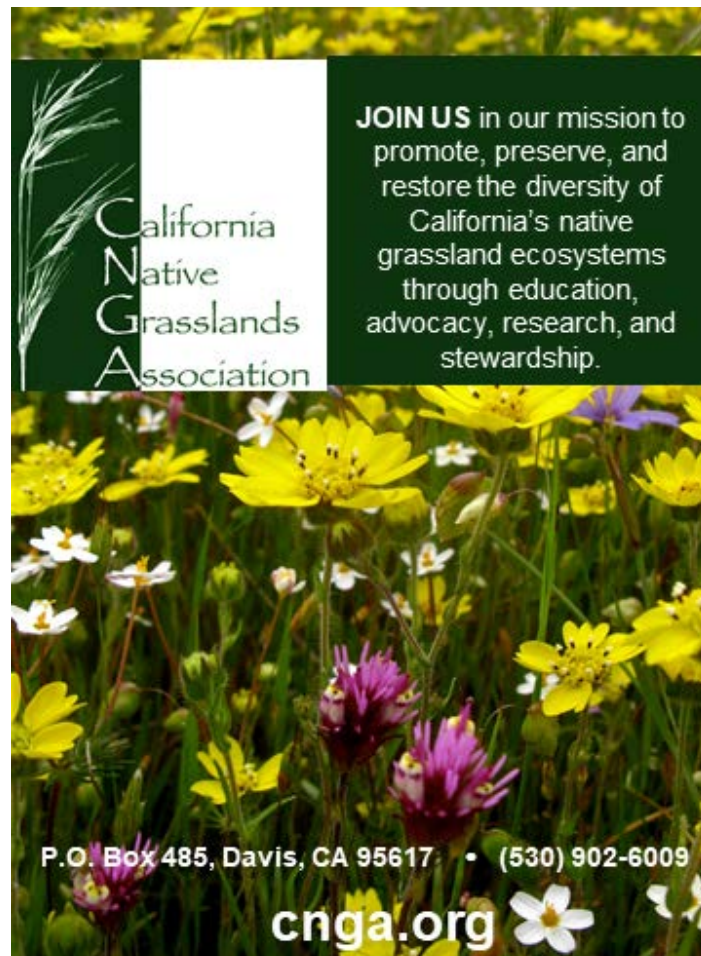
To test our hypothesis, we had to determine if a trend existed between biodiversity and 1) first flowering date, 2) peak flowering date, 3) or season length. We then compared the direction and strength of the trends between populations. The first analysis compared days to peak flowering of monoculture populations using an ANOVA test to confirm the relationship between latitude and flowering. We then determined if changes in days to peak flowering, days to peak flowering, and season length for each population were positive or negative in relation to biodiversity loss.

## Results

Data from the Santa Clara flowers, which represents the highest latitude population, were excluded from this first test due to low emergence rates, so only Riverside, Lost Hills, and mixed populations were compared. ANOVA results ( $F = 9.17$ ,  $p = 0.0038$ ) confirm that the peak flowering dates of the monoculture pots are statistically different (Fig. 3). The Riverside monoculture pots reached peak flowering earliest, then Lost Hills, and finally the mixed monocultures.

Figure 3 displays days to peak flowering while Figure 4 displays days to first flowering. Starting with the lowest latitude population, Riverside, days to peak flowering decreased as biodiversity increased and there was no discernible trend between biodiversity and days to first flowering. For the middle population, Lost Hills, days to peak flowering increased as biodiversity increased, while days to first flowering remained constant in relation to biodiversity. For the highest latitude population, Santa Clara, both days to peak flowering and days to first flowering decreased with biodiversity increase. However, this result is less reliable since the sample size was much smaller than other populations and no observations were made in the high biodiversity treatment. An interesting observation about days to first flowering was that the Lost Hills population flowered earlier than Riverside and Santa Clara flowers at every treatment level. Due to the lack of a consistent

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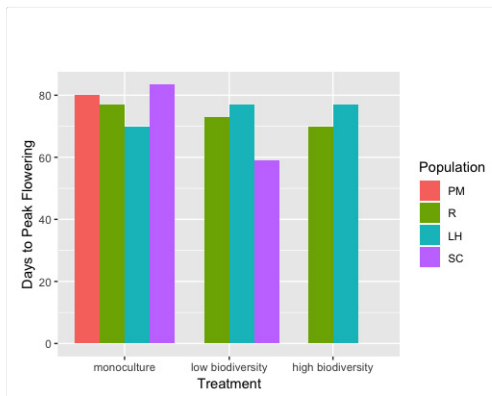


Figure 3. Mean number of days to peak flowering in response to biodiversity levels.

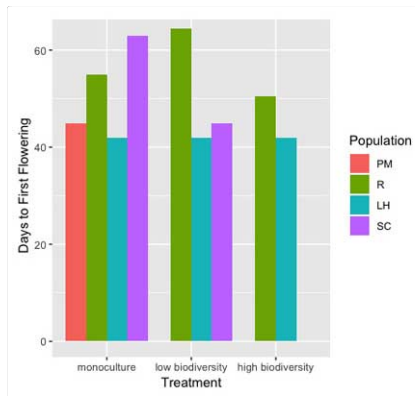


Figure 4. Mean number of days to first flowering in response to biodiversity levels.

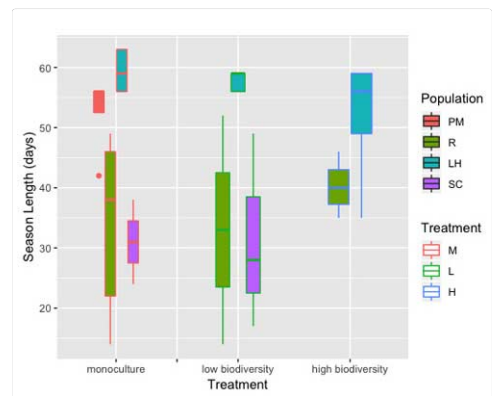


Figure 5. Flowering season length in relation to biodiversity levels.

## Biodiversity Loss and Phenology in California Grasslands *continued*

relationship between flowering and biodiversity, the trends observed within each population could not be compared to each other, and results of the mixed population treatment could not be compared to the other treatments.

Figure 5 displays the range of flowering season lengths for each population and treatment, measured by the average difference between the last and first flowering dates of each pot. The relationship between flowering season length and biodiversity is also inconsistent across populations. For Riverside, there is a slight positive relationship between season length and biodiversity. On the contrary, there is a negative relationship between season length and biodiversity in Lost Hills. There is an unclear flowering season length trend for Santa Clara. Additionally, the Lost Hills population and mixed population treatment group had much longer flowering seasons than either Riverside or Santa Clara. For all three measures, days to peak flowering, days to first flower, and flowering season length, there is no consistent pattern between population and response to biodiversity treatment.

### Discussion

The results of this study are inconclusive. Based on the collected data, declining biodiversity does not correlate with a consistent pattern in phenological response across the three needle goldfield populations. For all three measures, days to peak flowering, days to first flower, and flowering season length, there were both positive and negative relationships between biodiversity and phenological shift. With regards to the hypothesis, the expected direction of phenological shift in response to biodiversity loss occurred in only one of the three populations, Lost Hills, in which the days to peak flowering decreased as biodiversity decreased. However, the observed flowering season length in this population increased in relation to decreasing biodiversity, which is opposite to the expected direction of change and contradicts assumptions of the diversity-stability hypothesis. These two patterns occurred as predicted within the Riverside population: days to peak flowering increased and season length decreased in relation to biodiversity loss. Because of the conflicting results, the

original hypothesis cannot be confirmed or refuted, as there is no latitudinal trend connecting the opposing patterns observed. These results also suggest that the relationship between biodiversity and phenology, if any, may be more variable and complex within a species than previously thought.

While outside the scope of this study, inflorescence size differences were observed in relation to biodiversity levels. In general, inflorescences found in monoculture pots were larger in diameter (produced more flowers) than their counterparts grown in higher biodiversity environments. This trend could be indicative of excess competition. Although the total mass of seeds in each pot was standardized to minimize confounding effects of competition, the two grasses used to model California grasslands had such high germination rates compared to the needle goldfields that the low and high biodiversity pots were significantly more crowded than the monoculture pots. This seemed to result in increased competition, for nutrients, water, or sunlight, in biodiverse treatment pots. An area of further study would be determining whether a trade-off exists between inflorescence size and number in the presence of competition, as this would better inform ecosystem modelling.

The main limitations to this study are the small sample sizes both of needle goldfield populations and of replications. For one of the three populations, Santa Clara, germination rates were too low to comprehensively analyse flowering data across treatment groups. Many of the pots contained only one or zero needle goldfield plants. Due to a limited number of replications within each population, there was not enough data collected at each treatment level to run more statistical tests. Because Santa Clara was the only representative for high latitude populations, the lack of data also limited the ability to determine latitudinal trends. Future studies would benefit from more populations found along California's latitudes in order to account for differential emergence rates among populations and germination trials prior to experimental setup can reveal differences across populations. Alternatively, in studies with fewer sample areas available, replications

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# Biodiversity Loss and Phenology in California Grasslands *continued*

should be increased to capture more germination successes. Although the results of this study are inconclusive, the complex relationship between biodiversity and phenology should continue to be studied, especially in communities facing threats of biodiversity loss. The different patterns observed here pose a lot of questions regarding flowering patterns of needle goldfields, which will be informative to the conservation efforts in California grasslands.

## Acknowledgements

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