The effects of fire frequency on the reproductive success of buzz-pollinated blueberry (*Vaccinium pallidum*) and huckleberry (*Gaylussacia baccata*)

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Abstract

Fire suppression has dramatically modified wildlands in the United States. Following forest fires, the release of nutrients triggers flowering, potentially enhancing pollination services. Fire suppression and subsequent flower deficits may contribute to pollinator declines. The focus of our project is to determine the effects of fire on the reproductive success of two buzz-pollinated plant species, blueberry (*Vaccinium pallidum*) and huckleberry (*Gaylussacia baccata*). We predicted that fire would increase the reproductive success of these species through enhanced bee abundance. To test this, we quantified flower production and bee density along a fire gradient at Brookhaven National Laboratory, Upton New York. We estimated pollination success by quantifying the number and diversity of pollen grains on stigmas, and fruiting success. We found that the unburned site contained the most flowers, highest bumble bee density, and highest fruit set. Overall, bee abundances were low at the beginning of the year, potentially resulting in a decreased number of huckleberry and blueberry fruit sets this year

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compared to past years. Our results indicate that flower production, bee activity, and plant reproductive success were influenced by fire frequency, contrary to our hypothesis. Variation in weather may confound the effects of fire. 2016 was a particularly dry year with a warm winter, which may have dampened flowering and bee populations. Our results will assist Brookhaven National Laboratory’s Natural Resource Management Plan in the creation of a more efficient survey method addressing pollinator health.

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I.

~~I.~~ Introduction

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Fire suppression has the potential to cause a great loss of plant diversity and abundance in affected pine habitats. This method has been used by humans since the early 1900s, and causes the alteration of fire regimes throughout the United States [1]. Suppression turns pine habitats into closed-canopy forests dominated by oaks and other hardwoods and hinders the growth of many understory flowering ~~plants~~[plants [1].

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Pitch pine is globally rare, so it is important that we use active management to sustain it. Before management techniques were practiced, natural wildfires affected the Long Island pitch pine barrens. In 1938, following a period of frequent fires, 90% of vegetation in a study area was open canopy barrens. However, between 1938 and 1994, wildfires decreased in size covering less area, and as a result, barrens decreased to 45% of study area vegetation. Following this period, fires began to increase causing pine area to increase as well [2]. ~~2~~ ~~.~~

In order to maintain pine habitats, several common practices are the use of mechanical treatments, herbicides, and prescribed fires. Although herbicides and mechanical treatments are usually effective in eliminating unwanted plants, they can be toxic to wildlife. Prescribed fires avoid this issue, while being less costly. Fires release nutrients from organic matter and restart

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ecological succession, potentially increasing plant and wildlife diversity. Indeed, the abundance of understory plants is more sensitive to changes in management than the overall species richness, and plant diversity increased with intensity of disturbance [3]. Another study conducted by Clifford E. Lewis ~~[FIRST AUTHOR NAME]~~ and colleagues illustrated that vegetative growth and diversity increased in response to over 20 years of prescribed burning in a South Carolina coastal plain [4].

By ~~alterning~~altering landscapes, fire can influence pollinator services. The abundance, richness, and diversity of pollinators can be impacted by the habitat and landscape distributions of floral resources. Foraging ranges of *Bombus* species determine the size of the habitat utilized, and the area over which buzz-pollination of the host plants occurred [5]. Nesting resources such as sites and suitable soil for ground nesting bees could also be impacted by fire. However, “Potts et al. (2005) found increased abundance of ground nesting bees in recently burned areas due to increased bare ground available for nesting”[6]. Thus, fire can benefit pollinators as well, but its effects on insects have not been widely studied. A study on solitary bees conducted in the Mediterranean region found that burned areas were nearly absent of solitary bees due to direct mortality from fire or indirect from limited nectar sources (Ne’eman et al., 2000) [6].~~5.~~

The goal of this project was to survey the effects of fire on the reproductive success of buzz-pollinated blueberry (*Vaccinium pallidum*) and huckleberry (*Gaylussacia baccata*). We

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hypothesized that fire would increase flower production due to the release of nutrients and fruit through enhanced bee abundance and diversity. We tested these hypothesis in the pine barrens of Brookhaven National Laboratory along a fire gradient. To test these hypothesis, we quantified flowers, collected bees using netting surveys, examined pollen stigma loads, and counted bagged fruits.~~.~~

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II. Methods~~:~~

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A. Study location

This research was done in the pine barrens of Brookhaven National Laboratory from the end of May to mid-August in 2016. Within the Pine Barrens, three 15-acre sites were set up along a fire gradient (Fig. 1). One site was unburned, one site was burned in 2011 (once in the last 6 years), and the third site was burned in both 2011 and 2012 (twice in the last 6 years). There is also a fourth unburned site, but we did not use it for our data.

Figure 1. Map of the four study populations in the Pine Barrens of Brookhaven National Lab.

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B. Flower and bee density

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In order to monitor the direct effects of fire on flower production, we quantified density of blueberry (*Vaccinium pallidum*) and huckleberry (*Gaylussacia baccata*) flowers in each of 12, 0.5m x 0.5m subplots throughout the flowering season. .~~. ~~Due to misidenfication early in the summer, all flower counts were pooled across species. In order to determine bumblebee density, we conducted 30 minute netting surveys three times per week during blueberry and huckleberry flowering. All bumble bees (*Bombus* spp.) and blueberry bees (*Habropoda laboriosa*) were collected, identified to species and cast, and released. A subset of individuals were lethally collected for archival purposes. We tested for an effect of fire frequency on flower density and

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bee density via linear regression. A direct effect of flower density on bee density was via linear regression.

C. Pollination services and fruiting success

We quantified pollination services by surveying pollination success. We collected stigmas from one randomly selected blueberry and huckleberry near each subplot to determine the number and diversity of pollen grains on the stigmas. We counted the number of conspecific pollen grains (blueberry or huckleberry), pine pollen grains, and heterospecific pollen grains (excluding pine pollen) on each slide. Pollen was identified using a pollen library of all flowering plants in the area. Once fruiting commenced, we bagged one blueberry and one huckleberry plant containing fruit per plot at each site to protect against herbivory. Following fruit maturation, we collected each plant and counted the number of fruits on each, in order to test if fire has a direct effect on fruiting success beyond flower production. To test for a relationship between fire frequency and pollination success (i.e., conspecific pollen deposition, heterospecific pollen deposition, and fruit set), we conducted mixed-effect general linear models, with fire as a continuous, fixed effect and plant species as a random effect. We used general linear model with bee density as a continuous predictor variable and plant species as a random effect to test for a relationship between bee density and fruiting success.

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III. Results~~:~~

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A. Fire effects on flower and bee density

Fire had a negative effect on flower density ($F_{1,34}$ = 20.12, P< 0.0001, Fig. 2 ~~1~~), but not bee density ($F_{1,1}$ = 18.59, P = 0.15; Fig. 3A ~~2B~~). Bee density was correlated with flower density ($F_{1,1}$ = 161.28, P = 0.050, Fig. 3B ~~2A~~), indicating that fire has an indirect effect on bee density.

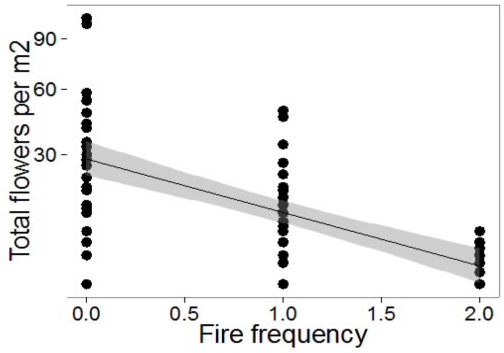

Figure 2~~1~~. Flower density was negatively correlated with fire frequency.

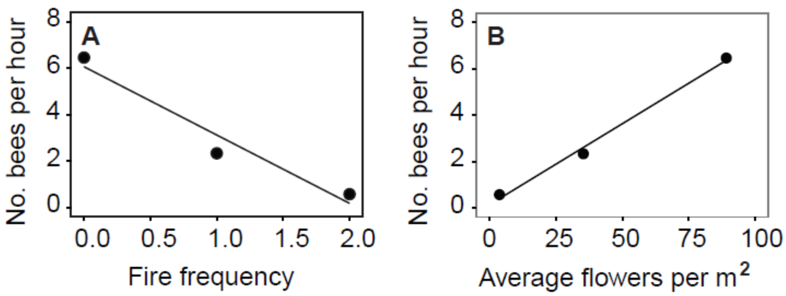

Figure 3~~2~~. Fire frequency (A) and flower density (B) influence bee density (number of bees collected per person hour).

B. Effects of fire on pollination services and fruiting success

Our preliminary analysis suggests that fire has a marginally significant negative effect on pollination success (number of confocal pollen grains on the stigmas) ($F_{1,18}$ = 2.70, P = 0.12; Fig. 4A ~~3A~~). While pine pollen was the most common type of foreign pollen, pine pollen abundance was highly correlated with abundance of non-pine heterospecific pollen grain deposition (t_{19} = 2.58, P = 0.019); therefore we pooled all heterospecific pollen types. There was no effect of fire on the amount of foreign pollen found on the stigmas ($F_{1,18}$ = 0.047, P = 0.83; Fig. 4B ~~3B~~), and no relationship between the amount of conspecific and foreign pollen present on the stigmas (t_{19} = -0.051, P = 0.96). Fire had a negative effect on fruit production ($F_{1,69}$ = 61.53, P< 0.0001; Fig. 5A ~~4A~~), likely mediated by pollinator abundance. As expected, bee density was positively correlated with fruit set ($F_{1,5}$ = 75.25, P = 0.0003; Fig. 5B ~~4B~~).

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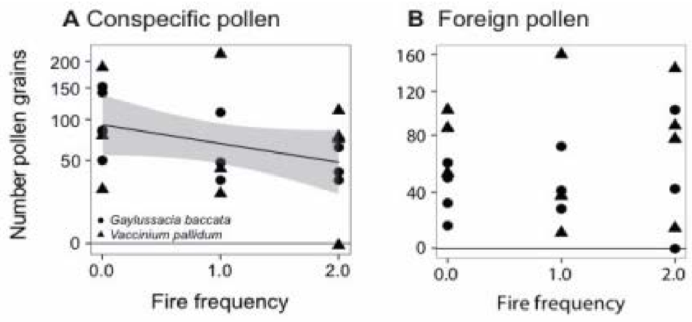
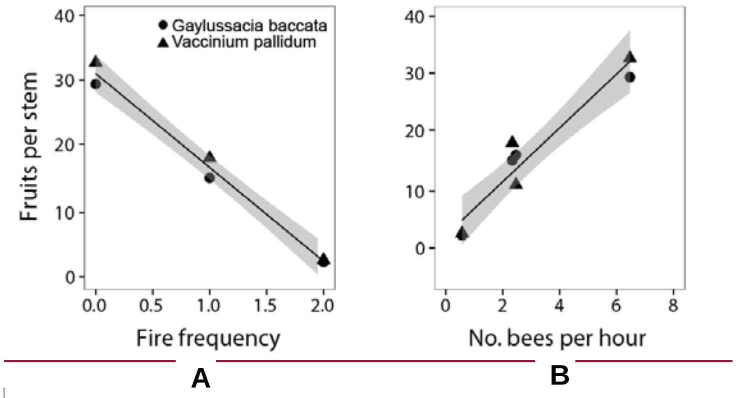


Figure 4~~3~~. Pollination success in two plant species, *G. baccata* and *V. pallidum*, along a fire gradient. The deposition of conspecific pollen (A) was related to fire frequency, but foreign pollen deposition was not (B).



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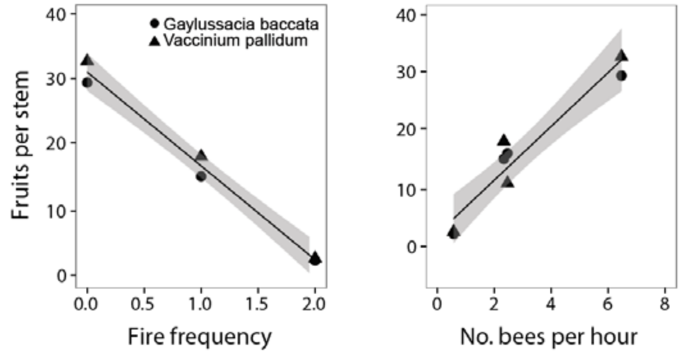

Figure 5~~5~~. Fruit production (number of fruits per stem) of *G. baccata and V. pallidum* was correlated with fire frequency (A) and bee density (number of bees per hour) (B).

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IV. Discussion~~:~~

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Our hypotheses that fire would increase flower production through nutrient release and fruiting success due to enhanced bee abundance and diversity were not supported. Our results indicate that fire had negative effects on fruiting and flower production. Although our data supports that fire has negatively affected plant and wildlife diversity in our sites, these declines in bumblebee population could potentially be influenced by climate changes. I~~While fire appears to have negative effects on fruiting success, i~~Interannual variation in precipitation may confound this relationship. There is significant evidence for this decrease in both domesticated and wild pollinators, as well as the plants that they rely on [7]~~[6]~~.However, in order to test this hypothesis, additional years of data would be needed in order to disentangle the effects of fire and climate.

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Since fruit set~~ was~~ positively correlated with bee density (Fig. 5B~~4B~~), it is possible that bee density was lower in the burned sites than the unburned site~~, since this is what our data showed~~ (Fig. 3A~~4B~~).

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However, we cannot assume that greater bee activity infers larger bee populations, because bumblebees can forage over long distances. Depending on the location of resources, bumblebees can forage up to distances of 1000 meters, with some exceptions if resources are extremely scarce [5]~~[5]~~. Hence, these bees could have foraged over greater distances in the Pine Barrens to access sites with more abundant food sources. Overall, our expectation that higher bee density would positively correlate with higher fruit set (Fig. 5B~~(Fig. 4B5A~~) and flower density (Fig. 3B)~~(Fig. 4A)~~ was supported, despite the fact that densities were higher in the unburned sites rather than the burned sites.

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Since there was no effect of fire on the foreign pollen found on the stigmas (Fig. 4B~~(Fig. 1B~~), and conspecific stigma loads were greater where fire was less frequent (Fig. 4A~~1A~~), we can infer that wind-pollinated plants may be less affected by fire than insect-pollinated plants. The increasing fire frequency indirectly~~negatively~~ affecting bee density (Fig. 3A~~4B~~) could have a

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direct impact on the decreasing flower density due to this relationship (Fig. 3B ~~4A~~). Information on wind-pollinated plants being less affected by fire than insect-pollinated plants is important, given the recent insect declines and prevalence of fire suppression. It is difficult to determine whether these population declines are directly impacted by fire suppression, or if outside factors such as climate change or pesticide use are causing this change.

Bees are declining worldwide [7]~~ADD REF~~]. Bees have exhibited shifts in population genetics, species level shifts [7], and bumblebee declines due to narrower climatic niches [7]. Climate changes affecting precipitation, soil humidity and snow melt (Inouye *et al.* 2003; Price & Waser 1998)[8] can disrupt the relationships among the environmental cues used by plants to initiate flowering, causing past combinations of cues to reappear at novel times throughout the season (Price & Waser 1998; Stenseth & Mysterud 2002; Visser & Both 2005)[8], resulting in bizarre flowering times [8]. This change poses a threat to pollinator habitats because it would affect their floral resource distribution.Climate change may also have indirect effects through phenological [7~~ADD REF~~] and morphological [~~ADD ~~Miller-Struttmann et al. 2015 Science] disruption of interactions between pollinators and plant species.~~ if they start responding differently, once they go through evolutionary changes.~~ ~~These~~

P~~P~~ollinator declines could have negative ecological and economic impacts that could significantly affect the maintenance of wild plant diversity, wider ecosystem stability, crop production, food security and human ~~welfare~~welfare [7]~~[6]~~. The findings of our study as well as similar studies can be used to sustain pollinator and plant diversity in the future.

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V. References

[1] ~~[1]~~Heuberger, K. A., and F. E. Putz. (2003). Fire in the suburbs: ecological impacts of prescribed fire in small remnants of longleaf pine (Pinus palustris) sandhill. *Restoration Ecology*, 11:72– 81.

[2] ~~[2]~~Jordan, M. J., W. A. Patterson, and A. G. Windisch. (2003). Conceptual ecological models for the Long Island pitch ~~pine barrens~~Pine Barrens: implications for managing rare plant communities. *Forest Ecology and Management*, 185:151–168.

[3] ~~[3]~~Griffis, K.L., Crawford J.A., Wagner M.R., Moir, W.H. (1 May 2001). Understory response to management treatments in northern Arizona ponderosa pine forests. *Forest Ecology and Management*, 146, 239-245.

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[4] [4]Lewis, C.E., Harshbarger, T.J. (January 1976). Shrub and Herbaceous Vegetation after 20 Years of Prescribed Burning in the South Carolina Coastal Plain. *Journal of Range Management*, 29, 13-18.

[5] [5]Geib, J. C., J. P. Strange, and C. Galen. (2015). Bumble bee nest abundance, foraging distance, and host-plant reproduction: implications for management and conservation. *Ecological Applications*, 25:768–778.

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[6] Campbell, J.W., Hanula J.L., Waldrop, T.A. (1 November 2006). Effects of prescribed fire and fire surrogates on floral visiting insects of the blue ridge province in North Carolina. *Biological Conservation*, 134:393-404.

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[7] [6]Potts, S.G., Biesmeijer J.C., Kremen, C., Neumann, P., Schweiger, O., Kunin, W.E. (24 February 2010). Global pollinator declines: trends, impacts and drivers. *Trends in Ecology & Evolution*, 25:345-353.

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[8] Hegland, S.J., Nielsen, A., Lazaro, A., Bjerknes, A., Totland, O. (28 November 2008). How does climate warming affect plant-pollinator interactions?. *Ecology Letters*, 12:184-195.

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VI. Acknowledgements

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I would like to thank Emma Squires and Emma Hert for their help in the field. This project was supported in part by the U.S. Department of Energy, Office of Science, Office of Workforce Development for Teachers and Scientists (WDTS) under the Science Undergraduate Laboratory Internships Program (SULI).