JOURNAL OF
EMERGING INVESTIGATORS

Article

Creating a phenology trail around Central Park Pond

Logan Flynn, Taran Pandya, Emilie Dion Wolf
The Browning School, New York, New York

SUMMARY

In this study, we aimed to determine whether the life cycle stages, or phenophases, of some plants in the urban environment of Central Park, New York, differ from the typical phenophases of the same plant species. To address this aim, we created the first phenology trail in Central Park, New York, by monitoring physical changes in thirteen plants over time and using DNA barcoding to confirm the visual identification of plant species on the trail. We isolated DNA barcodes from thirteen leaf samples by performing DNA extraction, PCR, and gel electrophoresis, before sending the samples for sequencing. We created an online map of our phenology trail using the program iNaturalist so that said map may be used by future researchers and citizen scientists. We hypothesized that the phenophases of the thirteen plants we studied would differ from their typical phenophases due to the 2 to 3 degree increase in temperature resulting from the urban heat island effect, which would have an impact on temperature-determined phenophases. We observed that, although the phenophases of five of our plants matched up with typical trends, there were distinct changes in the phenophases of the other eight, possibly resulting from the urban heat island effect.

INTRODUCTION

Phenology is the study of how seasonal variations and other environmental changes affect living organisms. Changing conditions affect the appearance of an organism and how it interacts with its environment. Biologists utilize phenological methods to observe the effects of such environmental changes on the natural world. Knowing about and observing such changes could help gather vital information that could impact environmental decision-making on local, national, and global scales. Phenology trails contain various organisms within their borders, which can be observed repeatedly in the same locations in an attempt to discern changes in the species (1). Since the eighteenth century, scientists and non-scientists alike have used phenology trails to observe and document the same organism over long periods of time (2-4).

Central Park is located in a metropolitan area that is known to be 2 to 3 degrees hotter than surrounding rural areas due to the urban heat island effect, creating a microclimate (5). The urban heat island effect results from an increase in heat energy in metropolitan areas compared to rural areas due to the high concentration of people, cars, and buildings in cities (6). The impact of an increase in temperature may vary between different plant species. In certain species, life cycle stages are determined by temperature, although light availability determines these same stages in others (7). Therefore, we predict that, due to the temperature increase from the urban heat island effect, the plants within the phenology trail will express different phenophases than typical members of their species.

This project sampled thirteen plant species that can be found within Central Park in New York. The species chosen for this project were the American yew (*Taxus canadensis*), ostrich fern (*Matteuccia struthiopteris*), common jewelweed (*Impatiens capensis*), Christmas fern (*Polystichum acrostichoides*), tulip tree (*Liriodendron tulipifera*), sassafras (*Sassafras albidum*), northern maidenhair fern (*Adiantum pedatum*), American holly (*Ilex opaca*), American pokeweed (*Phytolacca americana*), staghorn sumac (*Rhus typhina*), American common reed (*Phragmites australis subsp. americanus*), spotted lady's thumb (*Persicaria maculosa*), and Japanese maple (*Acer palmatum*). All but *P. maculosa* and *A. palmatum* are native to New York, meaning that the monitored *P. maculosa* and *A. palmatum* could have been cultivars that were planted in Central Park (8-10).

We posted photographs of the plants to iNaturalist, a website that allows observations and research to be shared amongst a social network of naturalists, citizen scientists, and other interested parties. iNaturalist utilizes photographs and descriptions provided by citizen scientists or naturalists to identify and monitor species around the world. This was incredibly useful for this project, as it allowed for effective data recording with species inquiries, observations, and locations around the Central Park Pond. Several of our photographs that were posted to iNaturalist have since been given the "Research Grade" status, confirming their species identity for our project and also allowing other scientists or citizen scientists to use them in their own research projects. Although we effectively sampled and collected data from all thirteen species, we were not able to definitively determine the identity of every species because of sequencing errors that most likely resulted from human error.

This project established the first phenology trail around The Pond in Central Park by collecting data on the phenology of thirteen plant species in the area. We aimed to ensure the continued use of this data by researchers and citizen

Table 1: Master table of species identification (visual and DNA) and DNA sequences

Visual Species Identity	Sample ID	Sequence Length	BLAST Result	Nucleotide Match	ID Quality
Matteuccia struthiopteris	OF	430 base pairs	*Matteuccia struthiopteris*	82%	Research Grade
Phytolacca americana	PB	571 base pairs	*Phytolacca americana*	87%	Research Grade
Liriodendron tulipifera	TT	567 base pairs	*Liriodendron chinense*	99%	Research Grade
Taxus canadensis	AY	570 base pairs	*Adiantum pedatum*	93%	Needs ID
Adiantum pedatum	N	572 base pairs	*Rhus typhina*	95%	Research Grade
Rhus typhina	SC01	653 base pairs	*Scomber scombrus*	89%	Needs ID
	SC02	552 base pairs	*Acer*	33%	Needs ID
Acer palmatum	JM	567 base pairs	*Taxus*	99%	Needs ID
Impatiens capensis	N/A	N/A	N/A	N/A	Research Grade
Polystichum acrostichoides	N/A	N/A	N/A	N/A	Research Grade
Sassafras albidum	N/A	N/A	N/A	N/A	Research Grade
Ilex opaca	N/A	N/A	N/A	N/A	Research Grade
Phragmites australis subsp. americanus	N/A	N/A	N/A	N/A	Research Grade
Persicaria maculosa	N/A	N/A	N/A	N/A	Needs ID

scientists alike. Over the course of this project, we found that eight of our study plants exhibited phenophase differences from expected trends over the eight month (October to May) study period. However, we were unable to definitively connect the impact of the urban heat island effect to these differences.

RESULTS

In order to identify our species, we collected DNA samples from the leaves of thirteen plants for DNA barcoding analysis. We were only able to collect samples from ten of our thirteen plants, of which only seven were successfully sent

Name	Sequence
OF-M13_A0.ab1	CNGCTATGACACNTCNCGNNGCCNATNTCTTTTGGATTCNNAGCTGGTGTCAAAGATTAC(
PB-M13_D0.ab1	TGTTGGANTTAAAGCTGGTGTTAAAGANTACAAATTGAATTATTATACTCCTGAGTATAAA
SC-M13_F0.ab1	TCAAAGCCGGCGTTAAAGACTATAAATTGACTTATTATACTCCTGAGTANNNNNNNNNNGI
TT-M13_C0.ab1	GGATTCAAAGCTGGTGTTAAAGAGTACAAATTGACTTATTATACTCCTGAATATNGAAAC(

Figure 1. Example nucleotide BLAST sequences for four study plants (OF - *M. struthiopteris*, PB - *P. americana*, SC - *R. typhina*, TT - *L. tulipifera*) (DNA Subway (11)**).**

off to Genewiz for sequencing, as the samples of *I. capensis*, *S. albidum*, *P. australis subsp. americanus*, and *P. maclosa* repeatedly failed to complete gel electrophoresis and the samples of *P. acrostichoides* and *I. opaca* did not undergo the extraction, PCR, and gel electrophoresis processes at all due to a lack of time for sequencing their DNA (**Table 1**). Some, such as *M. struthiopteris* and *P. americana*, were positively matched to the expected genus and species, while others, such as *L. tulipifera* and *P. australis subsp. americanus*, were only matched to genus level after sequencing, necessitating a search for visual clues in order to determine the exact species of the plants (**Figure 1**, **Figure 2**). Due to a handling error, the original sample of *R. typhina* was contaminated with fish DNA from a concurrent student project and was returned as *Scomber scombrus*, the Atlantic mackerel (**Figure 3**). In addition, due to another handling error that resulted in some of our samples being identified with the incorrect sample codes, we were unable to match sequencing results to the samples of *T. canadensis*, *A. pedatum*, *R. typhina*, and *A. palmatum*. Because of these errors, we identified these species through visual identification to compare the phenophases of our monitored plants and those of typical individuals of their species.

In addition to our DNA samples, we collected a series of photographs of each of our study plants, comparing each new photograph to previous ones to observe

#	Accession #	Details	Aln. Length	Bit Score	e	Mis-matches
1(1).	U05930.1	**Matteuccia struthiopteris** -	430	704	0.0	18
2(2).	JF832074.1	**Matteuccia struthiopteris** - Matteuccia struthiopteris ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene,partial cds	430	704	0.0	18
3(3).	KF186521.1	**Matteuccia struthiopteris var. pensylvanica** - Matteuccia struthiopteris var. pensylvanica voucher OAC 96872 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene,partial cds	430	704	0.0	18
4(4).	KJ841410.1	**Matteuccia struthiopteris** - Matteuccia struthiopteris voucher TJD-498 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds	430	704	0.0	18
5(5).	KJ593534.1	**Matteuccia struthiopteris** - Matteuccia struthiopteris voucher WAB_0132469103 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds	430	704	0.0	18
6(6).	KJ593533.1	**Matteuccia struthiopteris** - Matteuccia struthiopteris voucher WAB_0132469080 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds	430	704	0.0	18

Figure 2. Example nucleotide BLAST for *M. struthiopteris* (DNA Subway (11)**).**

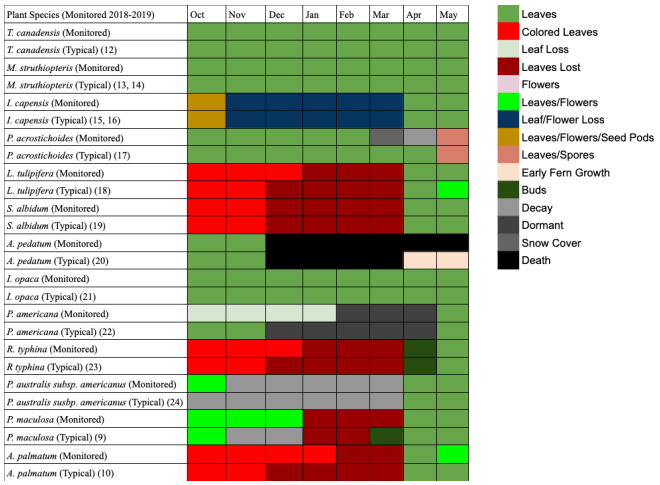

Figure 3. Phenophase comparison between typical trends and the observed phenophases during the course of the study.

phenological changes. From this data, we determined that the phenophases observed in eight of the thirteen target plants over the course of this eight-month study showed some deviations compared to those seen in the typical plants (**Figure 3**). The monitored *P. acrostichoides*, despite being an evergreen fern, had a brief period of decay in April, likely due to a thick layer of snow cover during the preceding March (**Table 2**) (17). The monitored *L. tulipifera* possessed leaves through December and did not possess any flowers during May, despite leaves typically being lost by December and flowers typically appearing during May (18). The monitored *A. pedatum* did die back in the winter, as is typical for the species, but never returned in the spring, possibly due to the removal of the plant by Central Park staff (20). The monitored *P. americana* continued to possess leaves through December before finally going dormant in January, despite the dormancy period typically starting in November (22). The monitored *R. typhina* continued to possess leaves through November despite leaves typically being lost during this month (23). The flowers on the monitored *P. australis subsp. americanus* were still growing in October, despite flowers typically disappearing by September (24). The monitored *P. maculosa* continued flowering through December and failed to bud in March, when flowers typically disappear in November and budding typically starts in March (9). Finally, the monitored *A. palmatum* possessed leaves through January and possessed flowers in May, despite leaves typically being lost by December and flowers typically not appearing until June or July (**Table 3**) (10). Although some of these deviations, such as those exhibited in the monitored *P. acrostichoides* and *A. pedatum*, are most likely the result of individual variation, others, such as those exhibited in the monitored *L. tulipifera*, *P. americana*, *R. typhina*, *P. australis subsp. americanus*, *P. maculosa*, and *A. palmatum*, suggest that the monitored plants within Central Park are exposed to conditions that impact which phenophases they express,

Table 2: Monitoring chart for *P. acrostichoides*

Monitored Characteristic	Height (cm)	Width/Diameter (cm)	Stage	Number of Leaves	Other Observations
October	N/A	N/A	N/A	N/A	N/A
November	20 cm	30 cm	Frond	6	Fronds flat and low to the ground, but still green in color
December	10 cm	20 cm	Frond	4	Fronds flat but still green, much of plant hidden by leaf litter
January	0 cm	8 cm	Frond	8	Green but lying flat
February	N/A	N/A	N/A	N/A	N/A
March	N/A	N/A	Snow Cover	N/A	Covered in snow
April	2 cm	50 cm	Frond	10	Growing in shade next to new fern growth, leaves still somewhat flat
May	50 cm	40 cm	Frond	18	Green fronds growing off the ground, small bumps on the underside of the fronds

although the specific identity of the environmental conditions that cause these phenophase changes remains unknown.

DISCUSSION

Although there are noticeable differences among the phenophases of our study plants from typical trends, there are not enough to say for certain whether these deviations are caused by the urban heat island effect. Some of our study plants, such as *T. canadensis* and *M. struthiopteris*, showed similar phenophases to those seen in typical members of their species, as their needles and fronds remained intact throughout the year (12-14), while other species, such as *P. americana* and *P. australis subsp. americanus*, showed some phenophase differences in regard to a dormancy period lasting from April to early May instead of from December to early May and atypical presence of flowers through early November, respectively (22, 24). Deviation between phenophases was more common during changes between seasons, such as the transition between fall and winter (October/November to December) and the transition between spring and summer (April to May), which are also

Table 3: Monitoring chart for *A. palmatum*

Monitored Characteristic	Height (cm)	Width/Diameter (cm)	Stage	Number of Leaves	Other Observations
October	N/A	N/A	N/A	N/A	N/A
November	N/A	93 cm	Colored Leaves	5,000+	Growing in a fenced in grassy field nearby The Pond, leaves turning reddish in color
December	N/A	93 cm	Leaf Loss	1-3 per branch	Little to no leaves present, branches intact
January	N/A	93 cm	Leaf Loss	0-1 per branch	Little to no leaves left, branches intact
February	N/A	N/A	N/A	N/A	N/A
March	N/A	93 cm	Leaves Lost	0	No leaves, growing in snow, no broken branches
April	N/A	93 cm	Leaf	3-5 on some branches	Some buds have begun to sprout leaves on some branches
May	N/A	93 cm	Leaf/Flower	7 per branch	Large, bright red leaves on every branch, some flowers present

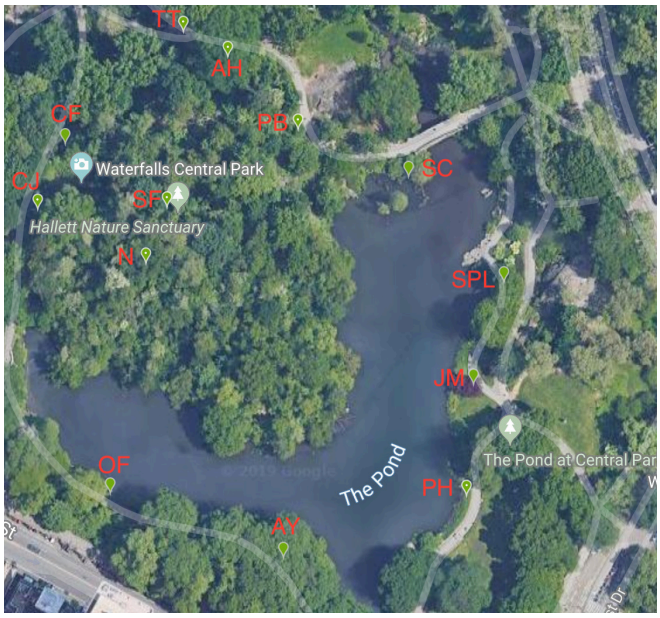

Figure 4. Map of the phenology trail around the Central Park Pond in New York (Green markers represent sample locations) (AY - *T. canadensis*, OF- *M. struthiopteris*, CJ - *I. capensis*, CF - *P. acrostichoides*, TT - *L. tulipifera*, SF - *S. albidum*, N- *A. pedatum*, AH- *I. opaca*, PB - *P. americana*, SC - *R. typhina*, SPL - *P. maculosa*, JM - *A. palmatum*, PH - *P. australis subsp. americanus*) (Google Maps (28)**)**

the periods during which the average temperature in Central Park tends to decrease and increase the most (**Figure 3**) (25-27).

The results of this study are significant since they are part of the first phenology trail in the Central Park area. This data can be used in the future to provide information about the status of these plants in the park at specific times and under specific conditions, which could be helpful for future environmental decision making. This data and all future data gained from this trail will continue to be used by researchers, citizen scientists, and Browning School students to observe the phenological changes on this trail long after the end of this study. In addition, several of our iNaturalist photographs have since been given the "Research Grade" status, confirming their species identity and also allowing other scientists or citizen scientists to use them in their own research projects.

Future studies that could originate from this project include a continued study of this phenology trail. After the end of this initial study, the phenology trail will remain intact and online for future use. Some experiments that could result from this trail include taking temperature recordings from the monitoring sites, which could help determine the impact of the urban heat island effect on phenological trends, or studying the impact of changing environmental characteristics on new and old growth plants. Other future experiments could include observing multiple members of the same species to determine whether or not environmental changes affect all individuals of the species in the same way or looking into various other factors, such as soil quality or access to water, to determine their impact on the observed phenophases. In addition, a complete year-round study of the trail could help determine whether the urban heat island effect has a stronger impact on the plants during certain months of the year, as this initial study did not collect observations during the summer months and had few monitoring periods during the months of October, December, February, and March. One limitation of this study that could be rectified by future experimentation is that the data on typical phenophases was not collected from the same time that the monitored plants were being observed. A future study could monitor individuals of the same species both on and off the trail at the same time in order to better determine differences between the monitored and typical phenophases. A final potential future study option could be the creation of further phenology trails in Central Park or in other areas of New York, such as Black Rock Forest, an area that is not influenced by the urban heat island effect, which could help determine whether or not the changes seen in this study are a result of the impact of this effect. These future studies are of extreme importance, as the results gained from them may help determine the effects of climate change on these and other species and may enable scientists to catch the early signals of these drastic effects and attempt to adjust policy accordingly.

MATERIALS AND METHODS

Thirteen species were selected along the trail surrounding the Central Park Pond, New York (**Figure 4**). The species were chosen based on their location and in order to have representatives of many different plant families. After being chosen, the species were visually identified by comparing images of the species to plant images collected by online databases such as iNaturalist and Nature's Notebook in order to find a match. The species ID was confirmed through iNaturalist with the help of scientists on the platform. Each plant was monitored as often as possible over a period of eight months, from October to May. Initially, the plants were monitored every two weeks, but monitoring sessions became weekly in April, as spring had begun and we wanted to see the rapid changes in each plant species. Each observation period consisted of measuring the height, width/diameter (taken at the widest part of each plant, including foliage), leaf number, phenophase, and any additional observations (**Table 2**, **Table 3**) (**Figure 3**). Species were photographed, and the data was recorded within the iNaturalist project "Central Park Pond Phenology Trail." This data was then compared with typical phenophases of the species, which were determined through data from outside researchers of various universities or organizations, in order to see how the phenophases of the monitored plants differed from typical trends. Physical samples of the plants were collected with permission from the Parks Department. A small piece of a leaf from each species was collected and placed in a plastic bag. The samples were refrigerated in the science lab at the

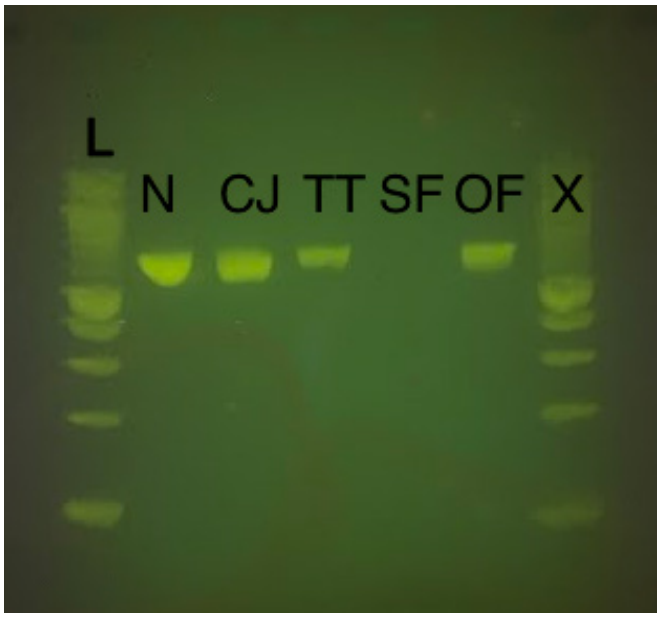

Figure 5. Example gel for five study plants (L - Ladder, N - *A. pedatum*, CJ - *I. capensis*, TT - *L. tulipifera*, SF - *S. albidum*, OF - *M. struthiopteris*, X - Unused Well**).**

Browning School.

The DNA was extracted and amplified following the PCR protocol outlined in the DNA Learning Center procedure, using the DNA Learning Center Barcoding Procedure Ready To Go PCR beads and *rbcL* primers, as *rbc*L is a gene found in the chloroplasts of most photosynthetic organisms (29-30). These samples were then loaded into a gel machine, and the resulting gel signatures were analyzed in order to ensure that the *rbc*L gene necessary for the DNA barcoding of these samples was present (see for an example gel **Figure 5**). Once the analysis was complete, the barcodes were sent to GeneWiz for sequencing, and DNA Subway was used to identify the species (11). The DNA barcodes were planned to be published to GenBank, but, due to the lack of accurate barcodes, none of them were sent for publishing.

ACKNOWLEDGEMENTS

We would like to thank the Browning School for providing us with the resources and funding that enabled us to perform this study. We would also like to thank Brady Simmons and the New York Parks Department for providing us with the permit to sample within Central Park, the Cold Spring Harbor Laboratory for providing funding, Christine Marizzi and the Harlem DNA Lab for providing us with the *rbcL* primers, and the Urban Barcoding Project for providing us with the extraction, PCR, and gel electrophoresis procedure.

Received: May 29, 2019
Accepted: March 7, 2020
Published: July 16, 2020

REFERENCES

1. Chicago Botanic Garden. “About Phenology.” *Budburst*, *Chicago Botanic Garden*, budburst.org/phenology-defined. Accessed 27 Sept. 2018.
2. BBC. “Robert Marsham: The Father of Springtime Records.” *BBC Norfolk*, BBC, 19 May 2008, www.bbc.co.uk/norfolk/content/articles/2008/05/01/nature_robert_marsham_20080501_feature.shtml. Accessed 12 Apr. 2019.
3. Nature’s Notebook. “Rio Grande Phenology Trail.” *Nature’s Notebook*, www.usanpn.org/nn/RioGrandePhenologyTrail. Accessed 27 Sept. 2018.
4. Nature’s Notebook. “Tucson Phenology Trail.” *Nature’s Notebook*, www.usanpn.org/nn/groups/tucson-phenology-trail. Accessed 27 Sept. 2018.
5. Southern Living Plants Collection. “What Is a Microclimate?” *Southern Living Plants Collection*, southernlivingplants.com/plant-care/article/what-is-a-microclimate. Accessed 12 Feb. 2019.
6. National Geographic. “Urban Heat Island.” *National Geographic*, www.nationalgeographic.org/encyclopedia/urban-heat-island. Accessed 12 Feb. 2019.
7. Li, Li *et al.* “Flowering Responses to Light and Temperature.” *Science China Life Sciences*, vol. 59, no. 4, 2015, pp. 403-408., doi:10.1007/s11427-015-4910-8.
8. U of Texas. “University of Texas Lady Bird Johnson Wildflower Center.” *U of Texas*, https://www.wildflower.org/. Accessed 21 Oct. 2019.
9. Tennessee Invasive Plants Council. “Polygonum persicaria.” *Tennessee Invasive Plants Council*, https://www.tnipc.org/invasive-plants/plant-details/?id=139 Accessed 21 Oct. 2019.
10. Bernheim Arboretum and Research Forest. “Japanese Maple.” *Bernheim Arboretum and Research Forest*, bernheim.org/learn/trees-plants/ bernheim-select-urban-trees/japanese-maple/. Accessed 30 Sept. 2019.
11. Cold Spring Harbor Laboratory. "DNA Subway." *DNA Subway*, Cold Spring Harbor Laboratory, dnasubway.cyverse.org/. Accessed 21 Sept. 2019.
12. U of Texas. "Taxus canadensis." *University of Texas Lady Bird Johnson Wildflower Center*, U of Texas, 10 July 2012, www.wildflower.org/plants/result.php?id_plant=taca7. Accessed 30 Sept. 2019.
13. U of Texas. "Matteuccia struthiopteris." *University of Texas Lady Bird Johnson Wildflower Center*, U of Texas, 7 July 2017, www.wildflower.org/plants/result.php?id_plant=MAST. Accessed 30 Sept. 2019.
14. Aderkas, P. Von, *et al.* "Leaf Development of the Ostrich Fern Matteuccia struthiopteris." *Botanical Journal of the Linnean Society*, vol. 93, no. 3, Oct. 1986, pp. 307-321., doi:10.1111/ j.1095-8339.1982.tb01028.x.
15. National Phenology Network. "Impatiens capensis." *Nature's Notebook, National Phenology Network*, mynpn.usanpn.org/npnapps/species/Impatiens/capensis. Accessed 30 Sept. 2019.

16. Schmitt, J., *et al*. "Density-dependent Flowering Phenology, Outcrossing, and Reproduction in Impatiens capensis." *Oecologia*, vol. 72, no. 3, June 1987, pp. 341-47., doi:10.1007/BF00377561.
17. U of Texas. "Polystichum acrostichoides." *University of Texas Lady Bird Johnson Wildflower Center*, *U of Texas*, 9 Sept. 2013, www.wildflower.org/plants/ result.php?id_ plant=poac4. Accessed 30 Sept. 2019.
18. University of Kentucky College of Agriculture, Food, and Environment. "Tulip Poplar." *University of Kentucky College of Agriculture, Food, and Environment*, *U of Kentucky*, www.uky.edu/hort/Tulip-Poplar. Accessed 30 Sept. 2019.
19. National Phenology Network. "Sassafras albidum." Nature's Notebook, National Phenology Network, mynpn.usanpn.org/npnapps/species/Sassafras/albidum. Accessed 30 Sept. 2019.
20. U of Texas. "Adiantum pedatum." *University of Texas Lady Bird Johnson Wildflower Center*, *U of Texas*, 4 Mar. 2016, www.wildflower.org/plants/result.php?id_plant=ADPE. Accessed 30 Sept. 2019.
21. Yale. "American Holly." Yale Nature Walk, Yale University, naturewalk.yale.edu/trees/aquifoliaceae/ilex-opaca/ american-holly-39. Accessed 30 Sept. 2019.
22. Florida Native Plant Society. "Phytolacca americana." *Florida Native Plant Society*, www.fnps.org/plants/plant/ phytolacca-americana. Accessed 30 Sept. 2019.
23. National Phenology Network. "Rhus typhina." *Nature's Notebook*, *National Phenology Network*, mynpn.usanpn. org/npnapps/species/Rhus/typhina. Accessed 30 Sept. 2019.
24. University of Maine. "Common Reed Fact Sheet." Signs of the Seasons, *U of Maine*, extension.umaine.edu/signs-of-the-seasons/indicator-species/common-reed-fact-sheet/. Accessed 30 Sept. 2019.
25. Sherry, Rebecca A., *et al*. "Divergence of reproductive phenology under climate warming." Proceedings of the National Academy of Sciences of the United States of America, vol. 104, no. 1, Jan. 2007, pp. 198-202., doi:10.1073/pnas.0605642104.
26. Ibáñez, Inés, *et al*. "Forecasting phenology under global warming." *Philosophical Transactions of the Royal Society*, vol. 365, no. 1555, Oct. 2010, pp. 3247-3260., doi:10.1098/rstb.2010.0120
27. National Weather Service. "Average Monthly & Annual Temperatures at Central Park." *National Weather Service*, https://www.weather.gov/media/okx/Climate/CentralPark/ monthlyannualtemp.pdf. Accessed 12, January 2020
28. Freeman, Scott. *Biological Science*. San Francisco: Pearson/Benjamin Cummings, 2008. Print.
29. Wolf, Emilie Dion, *et al*. Central Park Pond Phenology Trail Map. *Google Maps*, Google, 23 Sept. 2018, maps. google.con. Accessed 21 Sept. 2019.
30. Swink, Floyd A. "A Phenological Study of the Flora of the Chicago Region." *The American Midland Naturalist*, vol. 48, no. 3, Nov. 1952, pp. 758-68., doi:10.2307/2422209.
31. Urban Barcode Project. "Protocol for Uploading Gel Images of PCR Amplified DNA Samples." *Urban Barcode Project*, www.dnabarcoding101.org/files/Uploading-PCR-Gel-Images_2016-17.pdf. Accessed 28 Sept. 2018.
32. Diepstraten, Rianne AE, *et al*. "Does climate change and plant phenology research neglect the Arctic tundra?." *Ecosphere*, vol. 9, no. 9, Sept. 2018, pp. 1-11., doi:10.10002/ecs2.2362
33. Robert I. Bertin. "Plant Phenology And Distribution In Relation To Recent Climate Change," *The Journal of the Torrey Botanical Society*, vol. 135, no. 1, Jan. 2008, pp. 125-146., doi:10.3159/07-RP-035R.1