

BACKGROUND:

Climate change is impacting plant and animal communities, which can ultimately reshape the species, ecosystem services and forest management practices those communities support. Many plant and animal species are under threat from warming and must rapidly adapt through phenological shifts and/or range shifts northward to avoid harsher southern climatic conditions [Parmesan & Yohe, 2003; Schwartz *et al.*, 2006]. There is increasing evidence that climate change is exasperated at higher elevations [Giorgi *et al.*, 1997; Rangwala & Miller, 2012; Pepin *et al.*, 2015] and species' ranges could be restricted in these areas, potentially leading to regional extinction [Bachelet *et al.*, 2001; Potter *et al.*, 2008]. Thus, through the effects of stress and disturbance from warming, tree species' migration will be adversely affected, leading to profound impacts on forests and carbon sinks [Opdam & Wascher, 2004].

Natural forests are some of the most biodiverse habitats in the United States [White & Miller, 1988] and with climate change, the southeastern forests of Appalachia (Figure 1) are predicted to be under threat from increased wildfires and rapid conversion to savanna [Bachelet *et al.*, 2001]. Due to exploitative logging, clearcutting, grazing and wildfires at mid-elevations, these forests have become less complex over time, converted from historically mixed-oak stands to more homogenized stands of yellow poplar or red maple and American beech [Runkle, 1982; Lorimer, 1989; Rentch *et al.*, 2003a, 2003b]. Climate change coupled with rapid land-use change is resulting in the creation of gaps of varying size within forest canopies [Canham *et al.*, 1999]. These gaps within the canopy are introducing more microclimatic conditions within an ecosystem but the effects of these gaps on forest recruitment and resilience—or the ability of a forest to recover after a disturbance—are not fully understood and more research is needed.



Figure 1: Picture from research site in southern Appalachian forest in Asheville, NC.

Climate change is impacting forests in myriad ways—some of which are positive (i.e., increased CO₂ fertilization and longer growing seasons)—but many are detrimental such as increased stress from rising temperatures and decreasing precipitation leading to increased tree mortality from drought [Ayres & Lombardero, 2000; Bachelet *et al.*, 2001; Lloyd & Bunn, 2007; Allen *et al.*, 2010]. Repeated incidence of drought generally leads to increased vulnerability and subsequent decreases in forest resilience [Allen *et al.*, 2010; Anderegg *et al.*, 2020]. Understanding initial drought tolerance is therefore essential to predict future shifts in forest community dynamics. Some species will be more at risk of pests and pathogens following a drought and other habitats will have larger microclimatic variation, leading to a mosaic of drought risk within a forest [Ayres & Lombardero, 2000; Anderegg *et al.*, 2020]. By assessing both inter- and intra-specific variation in drought tolerance, pest damage and microclimatic impact, we can better predict the effects of climate change on temperate forests.

The combined effects of increasing temperatures and decreasing precipitation is impacting tree species differently, with extensive effects on drought-intolerant species leading to northward and westward range shifts [Fei *et al.*, 2017]. Additionally, there is growing evidence that southern Appalachian forests are transitioning to shade-tolerant, fire-resistant species such as red maple and American beech [Fei *et al.*, 2017; Knott *et al.*, 2019] and there is a reduction in foundation species' regeneration [Izbicki *et al.*, 2020].

Though oak species (i.e., *Quercus* genus) are generally fire-resistant, they are also shade-intolerant, thus forest management teams are working to regenerate oaks by establishing gaps in canopies in combination with prescribed fires. Recent studies suggest gaps must be large enough for oaks to regenerate successfully and demonstrate significant increases in photosynthetic rates and growing season lengths [Zhang & Yi, 2020]. Oaks are considered foundation species [Ellison

et al., 2005; Mitchell *et al.*, 2019] and greatly influence forest hydrology [Arthur *et al.*, 2012], nutrient cycling [Arthur *et al.*, 2012] and contribute to increases in biodiversity [Mitchell *et al.*, 2019; Izbicki *et al.*, 2020]. It is therefore essential to understand the effects of climate change on southern Appalachian forest habitats—with a strong focus on oak species—and the cascading impacts to our crucial carbon sinks.

Disturbance to canopy trees and the creation of gaps in forests can lead to a multitude of effects including increased competition through light availability as well as changes to soil temperature, moisture and microbial community structure. Canopy disturbance often leads to increases in soil nitrogen availability, which can allow for understory species to out-compete regenerating seedlings and saplings like oaks [Mladenoff, 1987; Taylor *et al.*, 2017]. Canopy gaps—especially more northern gaps, where sun angles are lower—with higher soil temperatures have significantly higher total growing season carbon flux than those with lower temperatures and less light availability [Raymond *et al.*, 2006; Schatz *et al.*, 2012]. Thus, identifying microclimatic soil variation in gap and closed-canopy sites is essential for accurate carbon flux forecasting and, by maintaining mixed-forest growth, there is a reduction in risk from the adverse effects of global climate change.

RESEARCH OBJECTIVES:

The overall aim of the proposed research is to understand how gap size impacts the reestablishment and maintenance of mixed-forest and regenerative growth. To do this I will investigate gap sites of varying size classes and compare these to closed canopy sites in the southern Appalachian Mountains to assess (1) forest recruitment of the dominant species and report diversity and richness of shade-tolerant vs shade-intolerant species over time, (2) drought tolerance of the dominant tree species across the gap and closed canopy sites using a greenhouse and phytotron cutting experiment and (3) soil microbial community structure, variability in soil nutrients, soil temperature, soil moisture and incident PAR of the gap sites versus the closed canopy sites **to understand and predict the impacts of climate change on temperate forest resilience.**

Hypothesis 1: The effects of gap size and location will impact species composition, recruitment and phenology. Using various gap types in comparison to closed-canopy forested sites in the southern Appalachian Mountains I will examine 10 different woody plant tree and shrub species—with overlapping phylogenies (Figure 2)—with 24 individuals per species: *Acer*

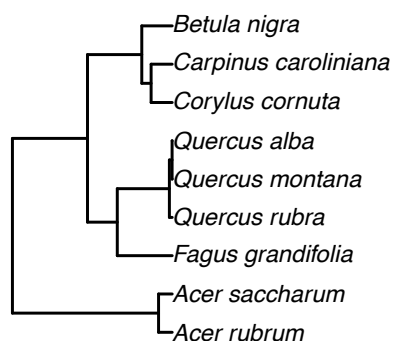


Figure 2: Phylogenetic tree of the focal tree species.

individual and also late season phenology (i.e., leaf drop and budset); and record carbon sequestration measurements. My experience in similar study investigating the effects of climate change across a latitudinal gradient will help prepare me for this work but I will also gain valuable guidance from Dr. Leggett, Dr. Keyser and Dr. Kalies.

Expected Outcomes and Significance: This experiment will greatly improve forecasts for mixed-forest, mid-elevation sites under climate change by developing equations and metrics of inter- and intraspecific variation in fitness to be used in climate models. I expect sites at the northern edge of large gap sites (i.e., gaps with diameter as large or larger than the height of the surrounding canopy trees [Raymond *et al.*, 2006]) will have longer growing seasons, warmer soil temperatures and greater carbon flux than closed-canopy sites. I also anticipate that mixed-forest, heterogeneous sites will have larger levels of recruitment and soil nutrients than more homogenized sites. Understanding the effects of warming—and the subsequent risk of disturbances—on temperate forests is essential for informing climate forecast models and determining forest resilience.

Hypothesis 2: Drought tolerance of the dominant tree species will vary across the gap and closed-canopy sites. Using a phytotron and greenhouse experiment, I will take cuttings from the focal tree individuals in Experiment 1 to test drought tolerance coupled with warming nighttime temperatures. Nighttime temperatures are increasing at a faster rate than daytime temperatures with warming [Fu *et al.*, 2016] and impact individual phenologies differently than daytime temperatures. There is also evidence for increased stress on plants from nighttime temperatures [Grinevich *et al.*, 2019]. We will additionally expose control and treatment individuals to varying increases in nighttime temperatures in the early season and during drought conditions. In the fall of 2021—after budset and before complete leaf drop, I will take 10-16 cuttings of approximately 30cm for each individual. Upon delivery to the lab, I will place the cuttings in dormancy conditions of 4°C for 10 weeks (Figure 3), rotating individuals every two weeks to minimize bias from possible phytotron effects. After 10 weeks, I will place the individuals in phytotron conditions and expose to 8 hour photoperiod and 15°C daytime temperatures and 10°C, 12°C and 15°C nighttime temperatures until leafout. Once full leafout is achieved, I will expose individuals to three levels of drought conditions: (1) control group, (2) little to no precipitation, (3) medium levels of precipitation, coupled with a full factorial experiment of increased nighttime temperatures (+0°C, +2°C and +5°C) for each drought group for a total of 9 treatments. Phenology, mortality, soil moisture, soil temperature and nutrient levels will be evaluated. After 8 weeks of drought conditions and increased nighttime temperatures, we will water half of the treatment groups to evaluate recovery by observing end of season growth through phenological measurements, canopy development, mortality and shoot apical meristem damage [Burgess, 2006; Blackman *et al.*, 2009; Brodribb & Cochard, 2009]. My experience with phytotron facilities running experiments assessing the effects of late spring freezing events on seedling and sapling development will help prepare me for this work but by working alongside Dr Doherty, I will hone my experimental skills in controlled environments, with a special focus on increasing nighttime temperatures.



Figure 3: Example of phytotron experiment

Expected Outcomes and Significance: By evaluating initial drought tolerance across the 10 dominant species of the southern Appalachians coupled with nighttime warming, I will be able to better predict the effects of increasing temperatures and decreasing precipitation from climate change on mixed-forest growth. I expect higher interspecific variability in drought tolerance and also low levels of intraspecific variation across the gap size and locations, with individuals from larger having higher levels of drought tolerance than closed-canopy individuals due to increased heat tolerance. In addition, I expect individuals exposed to higher nighttime temperatures will

demonstrate increased mortality and decreased recovery from drought. These findings are critical for forecasts as stress and disturbance are predicted to increase with warming.

Hypothesis 3: The variability in soil temperature, soil moisture and soil nutrients will increase with increased gap size. Understanding soil microbial community structure is a strong predictor for site response to environmental change. I will record hourly soil temperature at each site using Hobo Loggers buried 5cm below the soil surface and evaluate light availability using hemispherical canopy photos and then analyzing photos using Gap Analyzer software [Burton et al, 2014, Canham, 1990, Forrester et al., 2014]. Volumetric soil moisture will be measured monthly using a portable soil moisture probe and throughfall will be recorded for each field season. I will collect soil cores from 0-10cm and 10-20cm for each field season and compare to soil cores collected at the same or similar sites from 2017 (Dr. Leggett collected and archived samples in 2017) to compare soil microbial functional groups and nutrient content. I will then submit the soil cores to NCSU Soil Lab for standard nutrient analysis and to Microbial ID lab (Newark, DE) for PLFA analysis. Using structural equation modeling, I will evaluate the relationship of vertical and horizontal structure and soil microbial community structure. I have experience recording soil temperature, light availability and soil moisture but I will greatly develop my skills under Dr. Leggett's expertise in soil community structure and measurement techniques, which is essential for my career path in forest restoration as I have little experience in belowground ecosystems.

Expected Outcomes and Significance: Through the interactive effects of climate change and rapid land-use change, gap size and location will influence soil microclimatic conditions as well as nutrient availability. I expect light availability and soil temperatures to be greatest in the northern portion of the gap, while maximum soil moisture will occur in the southern portion of the gap [Schatz et al., 2012; Raymond et al., 2006]. By examining belowground responses to canopy gaps through soil moisture, temperature and nutrient composition, I will be able to greatly improve predictive climate models for the region and likely contribute to global modelling systems.

JUSTIFICATION OF SPONSORING SCIENTIST AND HOST INSTITUTION:

The proposed project and broader impact programs will be carried out in the lab of Dr. Zakiya Leggett in the College of Natural Resources at North Carolina State University. This project will allow me to transition to career independence and help broaden my skills from phenology observations and modelling [Chamberlain et al., 2020; Chamberlain et al., 2019; Ettinger et al., 2020; Furze et al., 2020] to studying ecosystem-scale processes---with a focus on both above and belowground systems---and forest resilience. In addition to developing my scientific skills, I will also leverage my leadership, professional development and diversity and inclusion initiatives. Throughout my graduate career, I primarily focused my research on investigating the effects of climate change on the intensity and frequency of late spring freezing events and the subsequent damage on forest ecosystems but I also helped lead a citizen science program and volunteered and worked part time for The Nature Conservancy. This experience has taught me how to work with different sectors and to teach and mentor individuals of all backgrounds and ages. Over the past several years, I have gained experience not only in publishing peer reviewed manuscripts but also documents for the general public and sharpened my teaching and communication skills with high school through graduate level students and the public. By joining the Leggett Lab, I will be able to further develop these skills while maintaining an interface with the nonprofit sector but I will also gain new research skills in soil carbon and nutrient cycling measurements. I have chosen to work with Dr. Leggett because my career goals are to work with BIPOC communities and, in the Leggett lab, I will have the opportunity to learn new, valuable teaching and mentoring skills---with a focus on BIPOC training---as well as expand my research and scientific toolkit to be more field based and incorporate more intensive field measurement skills.

Career Development: By working with Dr. Leggett at North Carolina State University, I will further develop my career path and work towards my goal of extending my research skills

alongside advancing diversity and inclusion at the university level. I will learn meeting facilitation, BIPOC training skills, grant writing, teaching and curriculum development as well as soil carbon and nutrient cycling measurement techniques. Dr. Leggett's background as an African-American female ecologist as well as her experience leading several programs to increase diversity and inclusion in ecology will make it possible for me to achieve my broader impact goals of learning techniques and strategies to increase and retain diversity in ecology while also maintaining my path towards understanding the effects of climate change on forests and our carbon sinks. This is a combination of skill sets that are unique to me working in the Leggett Lab group. In addition to working alongside Dr. Leggett, I will also work with Dr. Doherty (NC State University), Dr. Keyser (US Forest Service) and Dr. Kalies (The Nature Conservancy). My goal is to pursue a non-academic career in forest restoration and climate change mitigation with a special focus on environmental justice and inclusion of BIPOC communities. By working at NC State, I will be able to develop so many essential skills and tools simultaneously that will best prepare me for my personal, career aims.

PROPOSED RESEARCH TIMELINE:

Year	Semester	Research	Broader Impacts
2021	Summer	Identify focal individuals and plots Deploy hobo loggers Record observations for Exp 1 & 3	Develop IDP Recruit students for pipeline program Assist with citizen science study Co-facilitate UNIDE working group
2021	Fall	Record phenology; take cuttings for Exp 2	Develop and advertise course Begin mentoring and training students
2021	Spring	Set up Exp 2 Begin drought treatments for Exp 2 Record observations	Develop website for webinar series Teach course Maintain mentorship program
2022	Summer	Record phenology for Exp 1 Record observations for Exp 1 & 3	Begin webinar series Train students in field skills Supervise/advise undergraduates
2022	Fall	Record phenology Begin analyses	Update course materials
2023	Spring	Prepare manuscripts for submission	Offer the course again Maintain pipeline program to be useable in the future

BROADER IMPACTS

Teaching: Over the course of my proposed research timeline, I intend to develop and teach a 1-credit course for PhD students—especially BIPOC graduate students—how to develop postdoctoral grant proposals in ecology for NSF, USDA, NOAA or any other preferred agencies and assist students with securing a host advisor and/or university. By providing this opportunity, I will be able to hone my teaching and curriculum development skills while helping train students in an area not otherwise covered in the PhD experience. This course would involve bringing in guest lecturers and facilitating attendance to webinars offered by the aforementioned agencies. Under the Inter-Institutional Program with the University of North Carolina system, students from several other universities may register for courses at North Carolina State University. Therefore, this course could be advertised to doctoral students at other universities in the area including Duke, North Carolina Central University and University of North Carolina partner schools.

In addition to this course I will condense of the information gathered into a webinar that can be advertised to doctoral students nationwide. Dr. Leggett currently has a National Needs Fellowship

(NNF) program funded through USDA and has developed a collaboration with other NNF programs at University of Missouri and Michigan State University. All three of these programs in addition to ESA SEEDS and the Doris Duke Conservation Scholars program are developed to increase diversity in natural resources and ecology so the webinar will be advertised directly to these programs and any others similar to it. Additionally Dr. Leggett currently serves or has previously served on the advisory board at three HBCUs (Historically Black Colleges and Universities) that have doctoral programs and I will advertise directly to those programs. The webinar and the course will address the focal area of enhancing diversity at the postdoctoral level.

I will further sharpen my skills in meeting facilitation for ecology and natural resources by co-mentoring graduate students in the College of Education focused on a project entitled Natural Resources Diversity Curriculum Integration. The main project goal is to review entry level courses at North Carolina State University and develop modules to incorporate diversity and inclusion into these courses since they impact a large body of students early in their college experience. I will also co-facilitate working groups with an NSF funded RCN - The Undergraduate Network for Increasing Diversity of Ecologists (UNIDE). The project aims to build a sustainable and interdisciplinary network of ecologists, educators and social scientists to address how cultural and social barriers impact human diversity in ecology and environmental disciplines (EE). These two opportunities will help develop skills in the area of cross-sector engagement and meeting facilitation.

Mentoring: In addition to enhancing diversity at the postdoctoral level, I also propose to establish a mentorship pipeline for BIPOC individuals within the National Needs Fellowship (NNF) Program and the Doris Duke Conservation Scholars (DDCS) Program—where Dr. Leggett is lead PI and campus director. Both the NNF and DDCS programs have goals of recruiting and increasing diverse students in the field of ecology. Under the pipeline program I will develop, I will train graduate students, who will train undergraduate students who will in turn train high school students, with the goal of increasing diversity and interest in ecology at all levels. I intend to dedicate a large percentage of my \$15000 yearly fellowship allowance to funding the undergraduate and high school students in this pipeline program. By providing this training, I will better prepare students for postdoctoral work later in their career since they would have had this experience mentoring. Dr. Leggett is developing/building upon an existing training developed for the Doris Duke Conservation Scholars Program for mentors that have never advised BIPOC students for her NNF program, which will be used for the pipeline training program.

Community-level Outreach: In addition to my work with increasing diversity and inclusion at all levels, I intend to attend webinars and workshops offered by North Carolina State University designed for postdocs to hone project development and grant writing skills. I will also assist with grant writing to develop a collaborative project with The Nature Conservancy, which is working towards a project to evaluate forest carbon sequestration and increasing diversity and inclusion in program participants. I will also help Dr. Leggett develop research and education grants including expanding the National Needs Fellowship Program to include more students and developing a funding source for a dual degree program she has developed with Tuskegee University and North Carolina State University. Finally, I will work alongside Dr. Leggett, Dr. Asia Dowtin and Dr. Caren Cooper in the development and implementation of a large scale citizen science project which will involve students and communities across the nation that focuses on urban forestry/tree cover and BIPOC communities. Research has shown that BIPOC students are drawn to scientific pursuits that help local communities but deterred from disciplines that promote science as an isolating field [Puritty et al., 2017]. Therefore this citizen science project will provide an opportunity for these students and the participating public to nurture their interest in science while bringing awareness to their communities.