1. ADMINISTRATIVE:

Project Title: Forests of the Future: Integrated Assessment of Climate Change & Ecosystem Diversity

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Agreement number: G13AC00405
Date of Report: Dec. 19, 2017
Period of Performance: 2014 - Sept. 2017
Actual total Cost: $85,000

2. PUBLIC SUMMARY:

Understanding of the influence of global warming has been limited by a paucity of experiments. Taking advantage of the largest, longest-running experimental warming of a forest, we convened dozens of scientists from across the world to collect data to study and understand how bacteria, fungi, herbivores, plant pathogens, insects and a diversity of other groups respond to warming. We found that warming had a significant impact on ecosystems at both a site in North Carolina, as well as a more northern site in Massachusetts. The types of effects, however, differed between the north and south; they also differed as a function of the organisms considered. While warming affected all levels of organization, it had the greatest impact on phenology. Plants, microbes and animals were impacted to a similar extent, though the direction of the response varied by taxa, with some benefiting from warming, and other faring poorly. Warming impacted above ground responses to a greater extent than belowground responses. Overall, this experiment has provided us with a comprehensive picture of those taxa most likely to thrive or fail in light of the temperatures that will be experienced in the Southeast (and Northeast) in the next 100 years.

3. TECHNICAL SUMMARY:

Remarkably few experimental manipulations of the effects of climatic change, and in particular temperature change, have been conducted on animal populations and communities or on ecological processes dependent on their dynamics. This project built upon the longest-running, largest-scale forest warming experiment in the world to synthetically consider the effects of warming on diverse taxonomic and functional groups, from fungi and bacteria to herbivores and plant pathogens.

Prior to 2015, our work within the experimental chambers had focused on two main groups of organisms – ants and understory plants. Funds from the SECSC allowed us to expand the number of organisms whose responses to warming we considered through collaborations across North Carolina State University and other universities, both in the Southeast and globally. This included the study of snails, bacteria, fungi, herbivores, root biomass, soil composition, leaf-litter invertebrates (exclusive of ants), root nematodes, and other taxa. We were able to achieve this broad coverage through extensive collaboration using modern collection and sequencing techniques. The results represent the most ambitious attempt to understand how entire biotas change in response to warming conducted anywhere in the world to date.

4. PURPOSE AND OBJECTIVES: *This section should include information about the issue(s) the project addressed, and the community it serves. What were the original objectives identified during project initiation? Were they met? Have the original objectives been changed, eliminated, added to, or modified? Please describe any differences from the original proposal and why these changes were made.*

We had one overarching objective: to capitalize on the longest-running, largest scale forest warming experiment in the world to synthetically consider the effects of warming on diverse taxonomic and functional groups, from fungi and bacteria to herbivores and plant pathogens. We have addressed this objective well and more comprehensively than proposed in the project proposal.

As proposed, we convened a group of interdisciplinary scientists to create a collaborative plan for maximizing data collection and usage from the experimental warming chambers prior to shutting them down. The research leveraged the work and existing grants of a dozen other collaborators. We used sequence-based approaches to sequence the fungi, bacteria and, using metagenomics, arthropods present in each chamber. This generated a large body of data and analysis is ongoing and will likely generate new results for years to come as new analytical techniques are developed. Work on a large synthesis paper that summarizes results from 7 years of research is ongoing, as well as more than ten other papers that concern more in-depth analysis of individual taxa.

5. ORGANIZATION AND APPROACH: *This section of the memorandum explains, in task-oriented terms, how the research activities of the project were conducted. Briefly list which research methods were used to achieve results and why they were chosen by the team.*

We employed three core approaches. First, we convened meetings of interested faculty, postdoctoral researchers and graduate students from across the region in order to elaborate the approaches each would use to study their taxon of interest in the experimental chambers. Interest in the project was greater than initially expected, with over thirty scientists convening from across multiple institutions, states and countries to collect data on species and communities including plants, fungi, bacteria, and nematodes. Based on each collaborator's needs, we devised a sampling scheme and schedule and over the course of the following months, these collaborators collected data and samples from the chambers on a wide range of taxa, based on best practices in their own field of expertise.

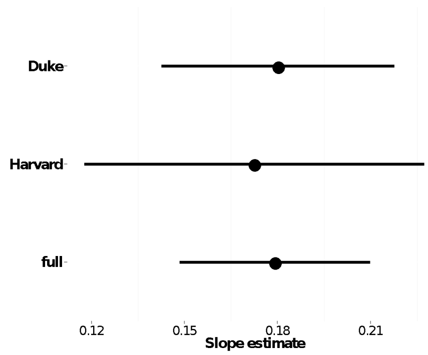
Second, we used sequence-based approaches to sequence the fungi, bacteria and, using metagenomics, arthropods present in each chamber. These samples have provided a comprehensive inventory for those samples of the taxa present including potential pathogens, mutualists, and even rare taxa. These data are being made freely available to all partners.

Third, given that the chambers provide a unique opportunity to study long term warming effects on a patch of forest, we wanted to ensure that we were maximizing the data we collected from the chambers. To this end, we collected and preserved a number of samples for potential future processing and analysis, even though there were no immediate plans to look at these samples by a specific collaborator. This included additional litter samples, soil samples, and ant specimen vouchers.

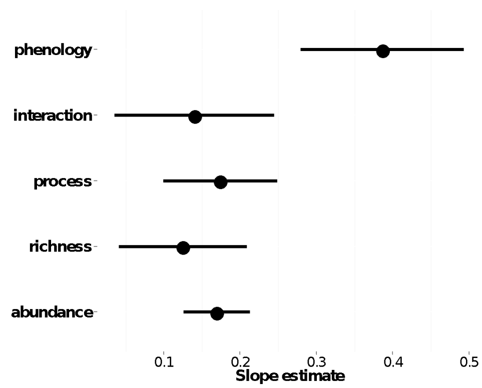
6. PROJECT RESULTS:

Work on a large synthesis paper is still ongoing due to the vast amount of data generated in the final year of the project. Below we have included some of the most striking and overarching preliminary results that we have to date.

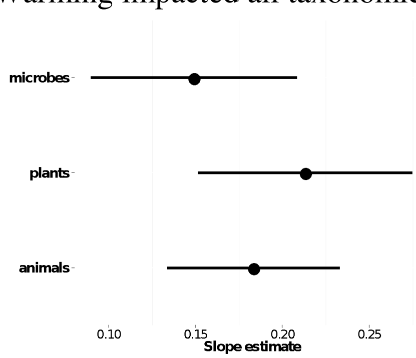
Warming had a significant impact on ecosystems, and this effect did not differ between sites (p = 0.26).



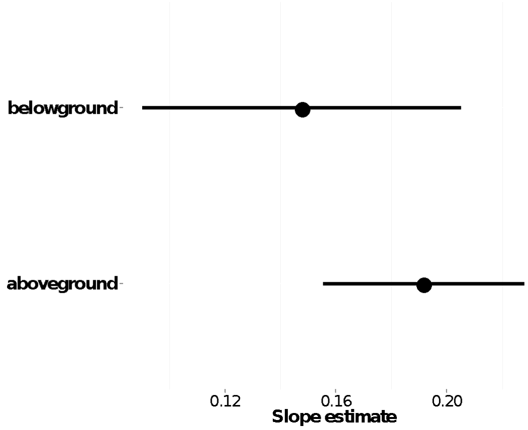
Warming affected all levels of organization, but this impact was significantly greater for phenology (p = 0.0006).



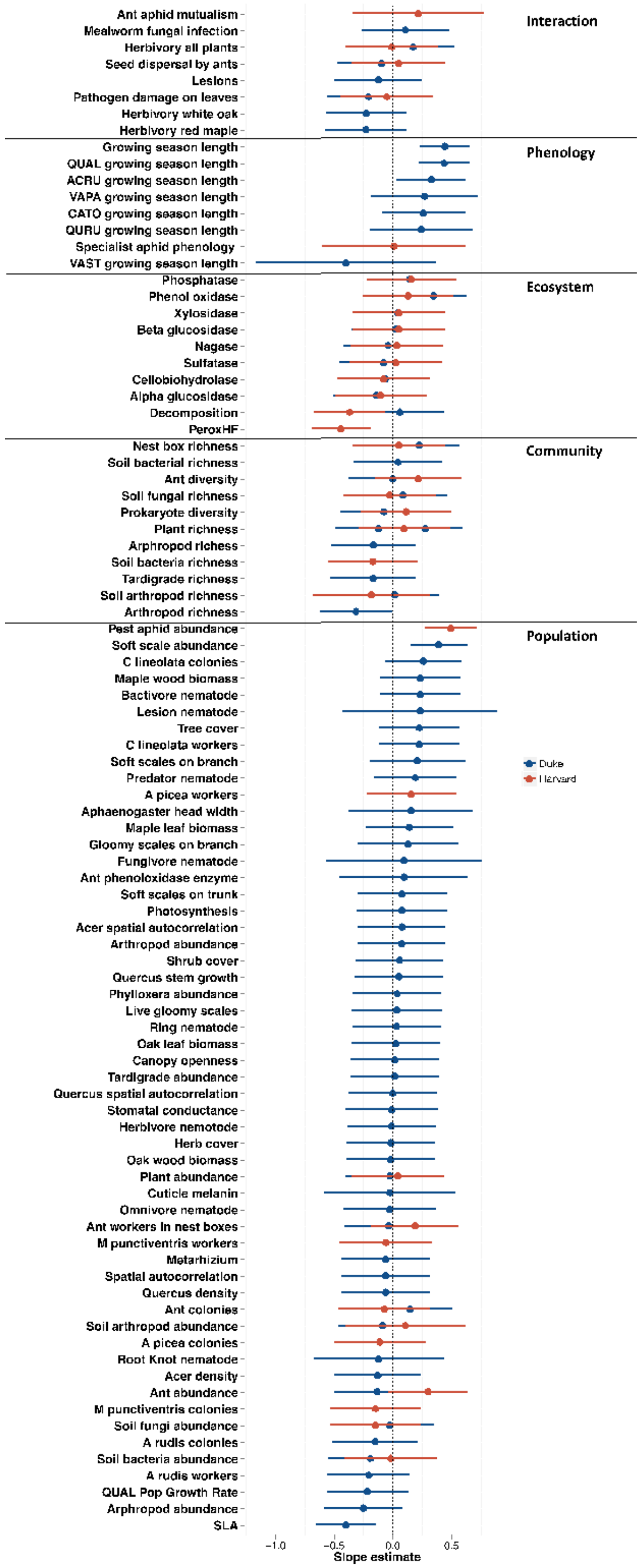
Warming impacted all taxonomic groups to a similar extent (p = 0.72).



Warming impacted aboveground responses to a greater extent than belowground responses, but the difference wasn't significant.



Responses to warming varied based on the variable measured. The figure below lists the ninety-two response variables, with effect sizes based on slopes in response to warming treatment:


Interaction
Ant aphid mutualism
Mealworm fungal infection
Herbivory all plants
Seed dispersal by ants
Lesions
Pathogen damage on leaves
Herbivory white oak
Herbivory red maple
Phenology
Growing season length
QUAL growing season length
ACRU growing season length
VAPA growing season length
CATO growing season length
QURU growing season length
Specialist aphid phenology
VAST growing season length
Ecosystem
Phosphatase
Phenol oxidase
Xylosidase
Beta glucosidase
Nagase
Sulfatase
Cellobiohydrolase
Alpha glucosidase
Decomposition
PeroxHF
Community
Nest box richness
Soil bacterial richness
Ant diversity
Soil fungal richness
Prokaryote diversity
Plant richness
Arphropod richess
Soil bacteria richness
Tardigrade richness
Soil arthropod richness
Arthropod richness
Population
Pest aphid abundance
Soft scale abundance
C lineolata colonies
Maple wood biomass
Bactivore nematode
Lesion nematode
Tree cover
C lineolata workers
Soft scales on branch
Predator nematode
A picea workers
Aphaenogaster head width
Maple leaf biomass
Gloomy scales on branch
Fungivore nematode
Ant phenoloxidase enzyme
Soft scales on trunk
Photosynthesis
Acer spatial autocorrelation
Arthropod abundance
Shrub cover
Quercus stem growth
Phylloxera abundance
Live gloomy scales
Ring nematode
Oak leaf biomass
Canopy openness
Tardigrade abundance
Quercus spatial autocorrelation
Stomatal conductance
Herbivore nemotode
Herb cover
Oak wood biomass
Plant abundance
Cuticle melanin
Omnivore nematode
Ant workers in nest boxes
M punctiventris workers
Metarhizium
Spatial autocorrelation
Quercus density
Ant colonies
Soil arthropod abundance
A picea colonies
Root Knot nematode
Acer density
Ant abundance
M punctiventris colonies
Soil fungi abundance
A rudis colonies
Soil bacteria abundance
A rudis workers
QUAL Pop Growth Rate
Arphropod abundance
SLA
Duke
Harvard
−1.0
−0.5
0.0
0.5
Slope estimate

Additional papers that have resulted from this work and the work of people funded by the grant include

Youngsteadt, Elsa, Andrew F. Ernst, Robert R. Dunn, and Steven D. Frank. "Responses of arthropod populations to warming depend on latitude: evidence from urban heat islands." *Global change biology* 23, no. 4 (2017): 1436-1447.

In this paper Elsa documents that the effects of warming on insects are contingent on background climatic conditions. The effects of warming are very different in warm regions than in cooler ones, in predictable ways.

Hamblin, April L., Elsa Youngsteadt, Margarita M. López-Uribe, and Steven D. Frank. "Physiological thermal limits predict differential responses of bees to urban heat-island effects." Biology letters 13, no. 6 (2017): 20170125.

Here, April shows that understanding the physiological tolerance of urban bees predicts their response to heat island effects, much as suggested by work in the climate chambers.

Dale, Adam G., and Steven D. Frank. "Warming and drought combine to increase pest insect fitness on urban trees." *PloS one* 12, no. 3 (2017): e0173844.

McCluney, Kevin E., Justin D. Burdine, and Steven D. Frank. "Variation in arthropod hydration across US cities with distinct climate." *Journal of Urban Ecology* 3, no. 1 (2017): jux003.

Meineke, Emily K., Anna J. Holmquist, Gina M. Wimp, and Steven D. Frank. "Changes in spider community composition are associated with urban temperature, not herbivore abundance." *Journal of Urban Ecology* 3, no. 1 (2017).

An additional ten papers are underway focused on specific components of the final project.

7. ANALYSIS AND FINDINGS:

While we initially intended to recruit local scientists to participate in our final data collection event, interest in the project extended far beyond the triangle area. Over thirty scientists from across more than ten institutions, seven states, and three countries came together to collect data from the chambers on a diversity of questions, from the microbial level up. Although sample collections have ended, we continue to create new collaborations and generate data and analyze the data in novel ways.

Based on our previous work in the chambers, we already knew that warming has a significant impact on ant diversity in forests and that this effect differed between southeastern and northeastern regions of the united states. We also knew that some species populations of ants became more common, while other populations of ants declined with warming, and that these shifts were due not only to direct effects of warming, but also to indirect effects of altered species interactions which in turn served to destabilize the ecosystem.

The collaborative effort of dozens of scientists is giving us a more cohesive view of how warming effects all organisms within an ecosystem, beyond just the ants. A synthesis of all of the research is ongoing, but from the preliminary analysis, we know that while ants responded more strongly to warming at our southeastern site, Warming had a significant impact on ecosystems as a whole at both sites. Warming also affected all levels of organization but it had the greatest impact on phenology, driven mostly by the growth in growing season for plants with warming. Plants, microbes and animals were impacted to a similar extent, though the direction of the response varied by taxa and variable measured, with some benefiting from warming (e.g. pest aphids, scale insects, acrobat ants, lesion nematodes), and other faring poorly (e.g. Aphaenogaster ants, metarhizium, red maples, overall arthropod abundance and richness). Though the difference wasn't significant, warming impacted above ground responses to a greater extent than belowground responses, suggesting that above ground processes might be particularly sensitive to warming, while those in the soil might be slightly buffered.

8. CONCLUSIONS AND RECOMMENDATIONS:

The project went as planned. In terms of recommendation for future work one of the clear take homes is that the effects of warming are extremely contingent on background climate. This contingency makes it difficult to imagine predictions that are fully general to all regions with regard to climate change. This may seem like a trivial conclusion, but it means that in practice each region needs to be studied independently, which is both costly and difficult. However, there are two hopeful generalizations that result from our work. One is that it does appear that one can make predictions as to how warm and cold climates will tend to respond to climate change and that such predictions are robust. In other words, one might be able to generalize within particular climate zones. The second, and perhaps more promising, is that our results from a long-term climate chamber largely match our results from the study of the long-term warming that has occurred in cities due to urban heat island effects. We currently have an invited paper from this project in review in TREE. This latter result is promising because it is far cheaper and more repeatable to study the effects of warming in urban contexts than through chamber experiments. In addition, whereas it is difficult to study some organisms in the context of chamber experiments (such as trees), it is easier in the context of urban warming. What is more, urban warming as an experiment, has been occurring over hundreds of years in some cities and just a few years in others such that it is possible to decouple long- and short-term effects.

9. MANAGEMENT APPLICATIONS AND PRODUCTS:

Management applications and products will await final publication of the main synthesis paper from this project.

10. OUTREACH:

Outreach has included a combination of peer-reviewed publications, conference presentations, site tours for school groups, and an active project webpage. The project has received media coverage from a number of media outlets, including National Geographic. Additionally, all data is being made public through an archive based at Harvard Forest and is accessible through the Harvard Forest webpage as well as the Dunn Lab project webpage listed above.

List of Peer-reviewed Publications:

Penick CA, Diamond SE, Sanders NJ, Dunn RR (2017) Beyond thermal limits: Comprehensive metrics of performance identify key axes of thermal adaptation in ants. Functional Ecology 31: 1091-1100.

Diamond SE, Chick L, Penick CA, Nichols LM, Cahan SH, Dunn RR, Ellison AM, Sanders NJ, Gotelli NJ. (2017) Heat tolerance predicts the importance of species interaction effects as the climate changes. Integrative and Comparative Biology 57:112-120.

MacLean, H.J., Penick, C.A., Dunn, R.R., Diamond, S.E. (2017) Experimental winter warming modifies thermal performance and primes acorn ants for warm weather. Journal of Insect Physiology. 100: 77-81.

Marchin, R.M., Broadhead, A.A., Bostic, L.E., Dunn, R.R., and Hoffmann, W.A. (2016). Stomatal acclimation to vapour pressure deficit doubles transpiration of small tree seedlings with warming. Plant, Cell & Environment 39, 2221–2234. doi: 10.1111/pce.12790.

Diamond, S.E., Nichols, L.M., Pelini, S.L., Penick, C.A., Barber, G.W., Cahan, S.H., Dunn, R.R., Ellison, A.M., Sanders, N.J., and Gotelli, N.J. (2016). Climatic warming destabilizes forest ant communities. Science Advances 2(10). DOI: 10.1126/sciadv.1600842.

Stanton-Geddes, J., Nguyen, A., Chick, L., Vincent, J., Vangala, M., Dunn, R. R., … Cahan, S. H. (2016). Thermal reactionomes reveal divergent responses to thermal extremes in warm and cool-climate ant species. BMC Genomics, 17, 171. http://doi.org/10.1186/s12864-016-2466-z.

Dale, Adam G., and Steven D. Frank. "Warming and drought combine to increase pest insect fitness on urban trees." *PloS one* 12, no. 3 (2017): e0173844.

McCluney, Kevin E., Justin D. Burdine, and Steven D. Frank. "Variation in arthropod hydration across US cities with distinct climate." *Journal of Urban Ecology* 3, no. 1 (2017): jux003.

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<u>In-prep:</u>

More than ten peer-reviewed publications are still in preparation and currently at the analysis stage. These include a large synthesis paper which will summarize the findings across all research conducted at the field site.