

Idiosyncratic responses of microbial mutualisms over 150 years of climate change

Joshua C. Fowler^{1,*}

Generic H. Collaborator²

Tom E. X. Miller¹

1. Rice University, Department of BioSciences, Houston, Texas 77006;

2. University of;

* Corresponding author; e-mail: jcf3@rice.edu.

Manuscript elements: Figure 1, figure 2, table 1, appendix A (for print; including figure A1, figure A2, and table A1), supplemental PDF. Figure 2 is to print in color.

Keywords: .

Manuscript type: Article.

Prepared using the suggested L^AT_EX template for *Am. Nat.*

Abstract

Lorem ipsum dolor sit amet, consectetur adipiscing elit. Sed non risus. Suspendisse lectus tortor, dignissim sit amet, adipiscing nec, ultricies sed, dolor. Cras elementum ultrices diam. Praesent quis dolor in dolor molestie cursus et ac nisi. Vestibulum ante purus, semper eget est vitae, vehicula ornare nisl. Morbi efficitur euismod enim, nec feugiat tellus cursus eget.

Introduction

Global climate change involves heterogeneous changes in multiple climate drivers [9]. It is of great interest to predict species' responses to these drivers. Novel conditions .Predicting how species' respond to these drivers, it is important to understand how and under what conditions interactions between species will alter expected responses. Incorporating interactions may lead to different predictions than those based on single species responses; for most species, we do not know whether their interactions will facilitate or hinder movement and adaptation to novel conditions [8]. Natural history collections, which were originally collected to study and preserve taxonomic diversity, present an exciting opportunity to explore long-term changes in ecological interactions across broad spatial scales [11]. These collections primarily comprise physical specimens of organisms collected along with information about the time and place of collection. These natural history collections, built and maintained by the contributions of thousands of scientists, are invaluable time machines. The specimens they contain are samples from dynamically changing environmental conditions through time and space. These specimens can preserve physical legacies of ecological dynamics and species' interactions. Recent work using collections has documented changes in phenology [3, 12, 22], as well as pollination [6, 13] and changes in negative interactions such as herbivory [10], but to date, focus has been lacking on long-term changes in reciprocally positive interactions, i.e. mutualisms.

Given that mutualists alter their partner's fitness, they may play an important role in limiting environmental stress, but changing conditions may also disrupt or breakdown the mutualism [2]. Examples of mutualism disruption as a result of changing climates include phenological mismatches between plants and their pollinators [13], drought-driven breakdown in seagrass-lucinid mutualisms [4], and coral bleaching due to temperature stress [17], all of which can have important consequences for habitat and biodiversity maintenance. Alternatively, mutualists may facilitate adaptive responses by improving fitness under stressful conditions, such as facultative bacterial symbionts within insects implicated in improving thermal tolerance [15, 19], or fun-

gal symbionts that allow their grass host to persist in more arid edges of its distribution [1]. Whether mutualisms breakdown or promote persistence under climate change is unclear, but likely depends on the intimacy and specialization of the interaction, as well as the physiological tolerances of the mutualists [14, 18, 20]. Given that climate change is spatially heterogeneous, the rate of change may also have a strong influence on when and where we can expect mutualism facilitation or disruption.

Early research on *Epichloë* fungal endophytes, symbionts of cool-season grasses, used herbarium specimens to describe the broad taxonomic diversity of hosts [21]. These endophytes are vertically-transmitted, growing throughout the aboveground tissue and into the seeds of their hosts, and consequently are preserved along with the plant on herbarium sheets. Vertical transmission links the fitness of host and symbiont together meaning that, over time, endophytes that provide fitness benefits should rise in prevalence within the population [5, 7, 16]. This leads to the prediction that interaction prevalence should generally be high where there are strong fitness benefits, and that prevalence should track changes in environmental drivers that limit fitness. Environmental stress is likely particularly consequential at species' range edges where population growth rates are expected to be near replacement levels []. And so understanding the ways that endophytes may mediate stress is important for understanding the factors setting species' range limits. The interaction prevalence should respond to the fitness effects of the mutualism, and the prevalence of endophytes across the range determines their actual contribution to setting range limits. In addition, the rate of environmental change is variable across space across species ranges [9], and local changes in endophyte prevalence likely depend on these rates.

And so, we are taking advantage of the spatial and temporal sampling within herbaria to ask how change in spatial and temporal environmental drivers alter the prevalence of a widespread mutualistic fungal endophyte symbiosis between *Epichloë* fungi and grasses. Specifically, we ask (1) how variation in temperature and precipitation correlate with endophyte prevalence across space and over time, and (2) does greater rates of environmental change drive greater changes in endophyte prevalence.

Methods

I will quantify the spatial and temporal responses of fungal endophyte symbiosis to environmental conditions by assessing the status of endophyte symbiosis in herbarium specimens. In particular, I will collect a small sample of seeds from herbarium specimens of *Agrostis hyemalis* and *Elymus virginicus* from across their distributions, and use microscopy to identify the endophyte status of specimens.

Second-order heading

Lorem ipsum nulla facilisi, pace ?]. Etiam semper, orci sit amet facilisis interdum, tellus nunc consequat erat, quis viverra nisi diam ut metus. Pellentesque cursus, sapien malesuada euismod iaculis, mauris purus interdum diam, vel vestibulum justo enim vitae tellus. Nunc interdum lorem sit amet diam volutpat tristique. Quisque pulvinar ac metus commodo lacinia (? ?).

Third-order heading

Usually two or three levels of heading will be all you need. Journal style even permits a fourth level in case you need it.

Fourth-order heading. The quick red fox jumps over the lazy brown dog in this paragraph as well. Donec mauris nibh, volutpat vehicula viverra at, iaculis congue sem. Praesent eget erat rhoncus erat sollicitudin volutpat.

$$\frac{1}{N_k - 1} \sum_{t=1}^{N_k} (M_{tjk} - \bar{M}_{jk})^2 \quad (1)$$

Praesent quis dolor in dolor molestie cursus et ac nisi. Vestibulum ante purus, semper eget est vitae, vehicula ornare nisl. Morbi efficitur euismod enim, nec feugiat tellus cursus eget. Donec mauris nibh, volutpat vehicula viverra at, iaculis congue sem.

Another second-order heading

As ?] argued, phasellus porttitor eros et ante condimentum, eget facilisis orci condimentum. Nulla facilisi. Proin placerat elit blandit, euismod dolor nec, dapibus diam. Mauris posuere malesuada lacus, at elementum lacus auctor eu (fig A1A).

Results

Lorem ipsum dolor sit amet. Aenean pulvinar malesuada commodo (see ? ; table 1). Sed aliquet mauris odio, in tristique dui egestas a. Etiam eu malesuada quam. Suspendisse tincidunt eu erat sit amet vulputate. Duis at arcu et nisl dictum mattis. Maecenas vel cursus ante. Cras eleifend elit nec velit sollicitudin fermentum in ac mauris. Pellentesque rutrum magna vel elit maximus hendrerit.

The height of the jump

Aenean eu pellentesque quam (fig. 1). Nam pellentesque augue eu finibus lacinia. Nullam nec justo vitae odio imperdiet rhoncus vitae vitae quam. Pellentesque porttitor metus et lectus ornare, ac cursus urna efficitur (fig A1B).

The laziness of the dog

Example paragraph with embedded references (video S1, fig. 2). If you have deposited data to Dryad, it is advisable to cite them somewhere in the main text (usually in the Methods or Results sections). A sentence like the following will do: All data are available in the Dryad Digital Repository (?).

Discussion

are a potentially rich target for study within museum specimens[]. Practically all specimens will be preserved alongside their microbiome, and while some evidence of these association can be expected to be lost over time due to specimen handling and storage.

Conclusion

Duis pharetra enim at libero cursus, eu commodo mi vestibulum. Nullam eget velit nec lectus viverra sodales. Suspendisse egestas, eros at dictum tincidunt, mi orci laoreet libero, eget rutrum sapien arcu blandit odio.

Acknowledgments

Statement of Authorship

Data and Code Availability

On initial submission, you may use this section to provide a URL for editors and reviewers that is ‘private for peer review’. After acceptance, this section must be updated with correct, working DOIs for data deposits (typically on the Dryad Digital Repository, ?) and code deposits (such as in Zenodo).

Appendix A: Additional Methods and Parameters

Fox–dog encounters through the ages

The quick red fox jumps over the lazy brown dog. The quick red fox has always jumped over the lazy brown dog. The quick red fox began jumping over the lazy brown dog in the 19th century

and has never ceased from so jumping, as we shall see in figure A1. But there can be surprises (figure A2).

If the order and location of figures is not otherwise clear, feel free to include explanatory dummy text like this:

[Figure A1 goes here.]

[Figure A2 goes here.]

Further insights

Tables in the appendices can appear in the appendix text (see table A1 for an example), unlike appendix figure legends which should be grouped at the end of the document together with the other figure legends.

Table A1: Various rivers, cities, and animals

River	City	Animal
Chicago	Chicago	Raccoon
Des Plaines	Joliet	Coyote
Illinois	Peoria	Cardinal
Kankakee	Bourbonnais	White-tailed deer
Mississippi	Galena	Bald eagle

Note: See table 1 below for further table formatting hints.

Lorem ipsum dolor sit amet, as we have seen in figures A1 and A2.

Literature Cited

- [1] Michelle E Afkhami, Patrick J McIntyre, and Sharon Y Strauss. Mutualist-mediated effects on species' range limits across large geographic scales. *Ecology letters*, 17(10):1265–1273, 2014.

- [2] Clare E Aslan, Erika S Zavaleta, Bernie Tershy, and Donald Croll. Mutualism disruption threatens global plant biodiversity: a systematic review. *PloS one*, 8(6):e66993, 2013.
- [3] Colette S Berg, Jason L Brown, and Jennifer J Weber. An examination of climate-driven flowering-time shifts at large spatial scales over 153 years in a common weedy annual. *American Journal of Botany*, 106(11):1435–1443, 2019.
- [4] Jimmy de Fouw, Laura L Govers, Johan van de Koppel, Jim van Belzen, Wouter Dorigo, Mohammed A Sidi Cheikh, Marjolijn JA Christianen, Karin J van der Reijden, Matthijs van der Geest, Theunis Piersma, et al. Drought, mutualism breakdown, and landscape-scale degradation of seagrass beds. *Current Biology*, 26(8):1051–1056, 2016.
- [5] AE Douglas. Host benefit and the evolution of specialization in symbiosis. *Heredity*, 81(6):599–603, 1998.
- [6] Yuan-Wen Duan, Haibao Ren, Tao Li, Lin-Lin Wang, Zhi-Qiang Zhang, Yan-Li Tu, and Yong-Ping Yang. A century of pollination success revealed by herbarium specimens of seed pods. *New Phytologist*, 224(4):1512–1517, 2019.
- [7] Paul EM Fine. Vectors and vertical transmission: an epidemiologic perspective. *Annals of the New York Academy of Sciences*, 266(1):173–194, 1975.
- [8] Sarah E Gilman, Mark C Urban, Joshua Tewksbury, George W Gilchrist, and Robert D Holt. A framework for community interactions under climate change. *Trends in ecology & evolution*, 25(6):325–331, 2010.
- [9] IPCC. *Managing the risks of extreme events and disasters to advance climate change adaptation: special report of the intergovernmental panel on climate change. Special Report*. Cambridge University Press, 2012.
- [10] Emily K Meineke, Aimée T Classen, Nathan J Sanders, and T Jonathan Davies. Herbarium

- specimens reveal increasing herbivory over the past century. *Journal of Ecology*, 107(1):105–117, 2019.
- [11] Emily K Meineke, Charles C Davis, and T Jonathan Davies. The unrealized potential of herbaria for global change biology. *Ecological Monographs*, 88(4):505–525, 2018.
- [12] Daniel S Park, Ian Breckheimer, Alex C Williams, Edith Law, Aaron M Ellison, and Charles C Davis. Herbarium specimens reveal substantial and unexpected variation in phenological sensitivity across the eastern united states. *Philosophical Transactions of the Royal Society B*, 374(1763):20170394, 2019.
- [13] Anton Pauw and Julie A Hawkins. Reconstruction of historical pollination rates reveals linked declines of pollinators and plants. *Oikos*, 120(3):344–349, 2011.
- [14] Nicole E Rafferty, Paul J CaraDonna, and Judith L Bronstein. Phenological shifts and the fate of mutualisms. *Oikos*, 124(1):14–21, 2015.
- [15] François Renoz, Inès Pons, and Thierry Hance. Evolutionary responses of mutualistic insect–bacterial symbioses in a world of fluctuating temperatures. *Current opinion in insect science*, 35:20–26, 2019.
- [16] Jennifer A Rudgers, Michelle E Afkhami, Megan A Rúa, Andrew J Davitt, Samantha Hammer, and Valérie M Huguet. A fungus among us: broad patterns of endophyte distribution in the grasses. *Ecology*, 90(6):1531–1539, 2009.
- [17] S Sully, DE Burkepile, MK Donovan, G Hodgson, and R Van Woesik. A global analysis of coral bleaching over the past two decades. *Nature communications*, 10(1):1–5, 2019.
- [18] E Toby Kiers, Todd M Palmer, Anthony R Ives, John F Bruno, and Judith L Bronstein. Mutualisms in a changing world: an evolutionary perspective. *Ecology letters*, 13(12):1459–1474, 2010.

- [19] Amy M Truitt, Martin Kapun, Rupinder Kaur, and Wolfgang J Miller. Wolbachia modifies thermal preference in *drosophila melanogaster*. *Environmental microbiology*, 21(9):3259–3268, 2019.
- [20] Robert J Warren and Mark A Bradford. Mutualism fails when climate response differs between interacting species. *Global Change Biology*, 20(2):466–474, 2014.
- [21] James F White and Garry T Cole. Endophyte-host associations in forage grasses. i. distribution of fungal endophytes in some species of *lolium* and *festuca*. *Mycologia*, 77(2):323–327, 1985.
- [22] Charles G Willis, Elizabeth R Ellwood, Richard B Primack, Charles C Davis, Katelin D Pearson, Amanda S Gallinat, Jenn M Yost, Gil Nelson, Susan J Mazer, Natalie L Rossington, et al. Old plants, new tricks: Phenological research using herbarium specimens. *Trends in ecology & evolution*, 32(7):531–546, 2017.

Tables

Table 1: Founders of *The American Naturalist*

Early editor	Years with the journal
Alpheus S. Packard Jr.	1867–1886
Frederick W. Putnam	1867–1874
Edward S. Morse	1867–1871
Alpheus Hyatt	1867–1871
Edward Drinker Cope ^a	1878–1897
J. S. Kingsley	1887–1896

Note: Table titles should be short. Further details should go in a ‘notes’ area after the tabular environment, like this.

^a Published the first description of *Dimetrodon*.

Figure legends

Figure 1: Figure legends can be longer than the titles of tables. However, they should not be excessively long—in most cases, they should be no more than 100 words each.

Figure 2: In this way, figure legends can be listed at the end of the document, with references that work, even though the graphic itself should be included for final files after acceptance. Instead, upload the relevant figure files separately to Editorial Manager; Editorial Manager should insert them at the end of the PDF automatically.

Figure A1: *A*, the quick red fox proceeding to jump 20 m straight into the air over not one, but several lazy dogs. *B*, the quick red fox landing gracefully despite the skepticism of naysayers.

Figure A2: The quicker the red fox jumps, the likelier it is to land near an okapi. For further details, see ?].

Video S1: Video legends can follow the same principles as figure legends. Counters should be set and reset so that videos and figures are enumerated separately.