

Decline for some, resilience for others: changes in the prevalence of microbial symbionts over 150 years of climate change

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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

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Introduction

Global climate change involves heterogeneous changes in multiple climate drivers [9]. Predicting species' responses to these drivers, whether species will move, adapt, or potentially go extinct is a major goal of basic ecological research and conservation management. Across the tree of life, species interact with macro- and microscopic symbionts which impact their growth and performance. These biotic interactions likely modulate responses to changing environmental conditions, yet for most species, we do not know whether they will facilitate or hinder movement and adaptation to novel conditions [8]. Predictions incorporating biotic interactions are likely different than those based on single species responses, but empirical assessments of these predictions have been limited by the lack of long-term and spatially extensive datasets of biotic interactions.

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-

luciniid 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 fungal 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 []. 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. We are taking advantage of the spatial and temporal sampling within herbaria to ask how the prevalence of a widespread mutualistic fungal endophyte symbiosis between *Epichloë* fungi and grasses has

changed in response to changing climate drivers. Specifically, the objectives of this study are to (i) quantify spatial and temporal change in endophyte prevalence, (ii) identify climate drivers that can explain changes in prevalence, and (iii) assess how the rate of environmental change influences changes in the prevalence of symbionts.

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

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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.

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

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Another second-order heading

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Results

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The height of the jump

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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 musuem 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

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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.

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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.