Ecological effect of evolution in pitcher plant rotifers as a response to temperature stress

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Rapid climate change has and continues to affect the health and trajectory of fragile ecosystems. Environmental stressors, such as increased annual temperature averages, pose a threat to many individual species by affecting their physiology and behavior. Given adequate genetic diversity, populations might have the potential to survive changes in their climate by evolving over short periods of time. Individuals that are more adept at living in novel environmental conditions are more likely to reproduce, and over an ecologically relevant timescale (a few to hundreds of generations) these advantageous traits may become ubiquitous throughout the population. This phenomenon of evolutionary rescue is well-studied in the field of evolutionary biology but paints an incomplete picture of what might actually be happening to the ecology of natural communities under these circumstances.

Traditionally, the study of evolution has focused on pairwise interactions between either a focal species and its environment, or a focal species and another community member—for example, a competitor, predator, prey, or mutualist. However, the effect that evolution in one species has on other members of its community may prove integral to how communities react to change. This interplay between evolution and ecology is known as eco-evolutionary dynamics. Recent studies have shown these microevolutionary processes play an important role in altering ecological effects and hold potential for expanding our understanding of ecology in natural communities. What is not well understood, however, is the extent to which stress-induced evolution has an impact on an organism's ability to compete with other community members. My goal is to examine how trait evolution over short time scales in response to stressful conditions associated with climate change may alter how organisms compete with other species in the community.

Although eco-evolutionary dynamics may be common in natural communities, observing such effects poses a challenge. A test for eco-evolutionary dynamics requires a controlled environment in which many generations of replicate populations can be observed evolving in response to stress and then quantifying the strength of interactions with other community members. Many aquatic invertebrates meet the first requirement—short generation times—meaning they hold the potential for measurable evolutionary change over many generations in only a few days or weeks. The bacterivorous (bacteria-consuming) rotifer *Habrotrocha rosa* is a microscopic aquatic invertebrate with a generation time of 1–3 days. In addition to this, *H. rosa* is obligately parthenogenetic and therefore incapable of sexual reproduction, meaning clonal cultures can be established from single individuals. Clonal cultures, consisting of thousands of genetically identical rotifers, enable the investigation of trait diversity in a population at the genotype level.

The ecology of this rotifer is also well suited for studying eco-evolutionary dynamics. *H. rosa* is found inside the cup-shaped rainwater-filled leaves of the purple pitcher plant, *Sarracenia purpurea*. The leaves of *S. purpurea* harbor a diverse community of organisms ranging several trophic levels. This type of micro-community, consisting of organisms inhabiting small volumes of water on or within a plant, is called a phytotelmic community. The conditions inside the phytotelmic leaf communities of *S. purpurea* can be replicated in laboratory microcosms that mimic ecological dynamics observed in nature.

Using the *S. purpurea* phytotelmic community, this project will address (1) the potential for microevolution in *H. rosa* by examining genotype-specific responses to stressful temperature, and (2) the effect of evolutionary change in *H. rosa* populations on interactions with competitors.

This summer, leaf water samples will be collected from *S. purpurea* in the Apalachicola National Forest. Thirty clonal families of *H. rosa* will be established, each seeded by a single rotifer. These cultures will be maintained in 50 mL centrifuge tubes and fed a natural community of bacteria.

Preliminary work has successfully resulted in the isolation of 20 distinct clones of *H. rosa* from samples provided by a collaborator at Florida State University. In the coming weeks, I plan to determine the extent to which trait diversity exists both within and among pitcher plant leaves using the samples I have available. Results from these preliminary experiments will provide data essential to the final design of the following experiments.

(1) To investigate the potential for microevolution in *H. rosa,* different genotypes (n = 30 clones) will be exposed to stressful temperature. Each experimental culture will be inoculated with cohorts of 50 rotifers from a single clone. Cultures will be replicated three times per treatment per clone and exposed to one of two temperatures: ambient (25°C) and stressful (37°C)—the latter of which is predicted by near-future climate projections in the southern range of *S. purpurea*. Abundance will be measured daily to estimate population growth rate. After 15 days of temperature exposure, the mass and respiration rate of a sample of individuals from each replicate will be measured. I expect clones to respond differently to increased temperature, indicating potential for selection on these traits in pitcher plant communities. This affects not only the fitness of the rotifers themselves, but may also affect other species with whom they compete.

(2) In a separate experiment, 10 mixed cultures of all clones will be exposed to ambient or stressful temperature for 2 months, during which selection for particular clones, and thus evolution, can occur. A sample of the final population from each of these mixed cultures will then be exposed to a fully crossed experiment including both stressful or ambient temperature, and the presence or absence of the bacterivorous protozoa *Tetrahymena thermophila*. This protozoan is a single-celled competitor that *H. rosa* is likely to encounter in nature, and the presence or absence of this competitor will be used to observe differences in the competitive ability of *H. rosa* populations in light of their evolutionary histories. Size, population growth rate, and respiration rate of each population will be measured post-exposure after a period of 1–3 generations to avoid phenotypic plasticity—a common confounding factor in selection experiments. Selection for traits that are more successful in stressful temperatures may alter the competitive ability of populations of *H. rosa*. A difference in competitive ability would indicate an ecologically relevant impact of microevolution.

The set of traits that are more fit at higher temperatures may be associated with a different ability to compete with other bacterivorous community members, like the protozoa *T. thermophila,* Evolutionary rescue could cause a population of rotifers to be better or worse at competing with other community members. Because of this, considering eco-evolutionary dynamics may prove essential to the conservation and restoration of fragile natural communities as they experience climate-induced environmental change.