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Temperature and Nutrient Change and their Evolutionary Consequences

Adam Pellegrini, Class of 2010

Environmental stress can cause populations to undergo differential evolutionary change. This evolutionary change can be a function of the intensity of the stress, other associated abiotic/biotic interactions, and species specific ontogenic factors. This study investigated the effects that temperature stress (5°C increase) combined with varying nutrient allocation has on two epiphytic bromeliad species, Vriesea cathyi and Tilandsia bulbosa on biomass production and growth rates over 40 days. I hypothesized that plants under high temperature would have a lower growth rate and lower biomass than control plants and that different nutrient levels will cause significant differences in biomass production and growth rate. Plants in the cooler temperature produced more relative biomass, specifically in the Phosphorus treatment in Tilandsia bulbosa, and had higher relative growth rate in both species. There was a significant effect of nutrient treatment (Nitrogen-Phosphorus treatment had the lowest growth) in the 27°C Tilandsia bulbosas and in the 27°C and 32°C Vriesea cathyi. There was not a significant effect of temperature on growth rate in the Tilandsia bulbosa but there was a significant effect of temperature on growth rate in Vriesea cathyi. Differential nutrient availability did not affect either an epiphyte's response to temperature. Within the significant effects of both treatment and nutrients, I found different effects across species. For example, while the relative biomass data patterns were consistent across species, the relative growth rate were not, with new leaves in the V. cathyi growing significantly faster than new leaves in T. bulbosa. This study showed that temperature change affects epiphytes, a particularly vulnerable plant group, more so than nutrient levels changing.

Introduction

The ability of an organism to cope with environmental change depends on its ability to respond to this change. This response can be through either a physiological change (phenotypic) or by moving to a new habitat that is similar to the organism’s old environment (dispersal). As the Intergovernmental Panel on Climate Change has projected a minimum of 5°C Celsius increase over the next century and an even greater maximum increase (IPCC 2007), it is necessary to investigate how this temperature increase will affect abiotic and biotic interactions. An indirect effect of temperature increase is the increase in evapotranspiration (ET), which results in there being greater net water loss within a plant. Although net primary productivity is positively correlated to ET (Lieth & Whittaker 1975), a system that is experiencing a greater net water loss might not see this positive correlation between NPP and ET. Though this correlation between NPP and ET is significant, RGR also depends on initial plant size as well as ET (Laube & Zotz 2003).
In plants, multiple nutrient uptake relationships and respiration rates depend on temperature and CO$_2$ levels (Ryan 1991); however, the data on how plants will respond to temperature changes is not clear-cut. Epstein et al. (2000) found that in Arctic plant communities increased temperatures caused an increase in nitrogen mineralization rates and growing season, which resulted in increased biomass. Contradicting Epstein, other studies have shown that when growth temperature was increased by 3°C, final plant biomass was reduced by 30% (reviewed by Zotz & Bader 2009). Shaver (2000) also showed that not all ecosystems will respond the same way to rising temperature.

In addition to temperature, nutrients greatly regulate the amount of NPP of a plant (Wedin & Tillman 1996). The majority of plants are Nitrogen (N) limited, especially when the N:P ratio is below 16 (Koerselman & Meuleman 1996). Studies have been focused on the abundance and mobility of N and Phosphorus (P) in soils and how this affects which nutrient is limiting. Epiphytes primarily undergo nutrient uptake through their leaves. More generally, plants respond to nutrient concentration by varying their biomass production. Nutrient use efficiency (NUE) and RGR are the main measures for how plants allocate nutrients to biomass. NUE is maximized when plants have high resorption efficiency, enabling the plants to recycle nutrients (reviewed by Aerts & Chapin 2000). Chapin et al. (1986) defined one measure of nutrient limitation to be the “enhancement of primary production in response to a large addition of the limiting nutrient.” Empirical studies can use this measure to determine important nutrient uptake characteristics of plants. Multiple factors, including elevated CO$_2$ levels (Bassititad 2000) and plant size (Reich et al. 2006) are positively correlated with N uptake and therefore affect NUE, SLA, and RWR. All of these studies display the complexity of nutrient uptake and the necessity of measuring a wide range of variables.

Most of these studies on temperature and nutrient change have focused on land plants. Epiphytes, which are plants that live on plants, are in the interchange zone between the atmosphere and the biosphere; thus, they may be especially sensitive to global change (Zotz and Bader 2009). Additionally, there are approximately 25,000 species of vascular epiphytes (Benzing 1990), making them a crucial plant group to study. Nutrient and temperature change may affect epiphytes differently because Epiphytes, unlike the majority of land plants, are theorized to be limited not by nutrient levels, but by water (Zotz & Heitz 2001), which may make the role that ET plays of NPP even greater. The physiological variance in epiphytes allows certain morphology to tolerate water stress better (tank bromeliads) or worse (bark bromeliads) (Benzing 1990). It is currently unclear as to the direct effects that nutrient limitation has on epiphytes because of the corresponding high stress of the instability of water supply (Zotz & Hietz 2001). This high uncertainty as to what governs epiphyte growth combined with their associated high diversity shows that a study into the effects of how combined temperature and nutrient levels has on
plant growth. Because of the complex relationship between temperature and nutrient levels governing plant growth, this study used a factorial design to explore how a 5 degree C temperature increase will affect growth. I measured the effects of increased temperature on the nutrient uptake dynamics in two species of bromeliads (*Vriesea cathy*, *Tilandsia bulbosa*). Bromeliads are vascular epiphytes, which are plants that live on other plants (see Cardelús et al. 2006). *Vriesea cathy* is a tank bromeliad that has the potential to store water for an extended period of time in its phytotelmata, while *Tilandsia bulbosa*, an atmospheric bromeliad, has a much more limited time for moisture uptake. I hypothesize that 1) there will be a significant decrease in growth (both as a function of biomass and rate) in the plants exposed to the higher temperature. 2) There will be a significant effect of nutrient treatment on growth.

**Methods**

**Plant Species**

My study species were *Vriesea cathy* and *Tilandsia bulbosa*. Both species are native to lowland rain forest and were propagated in Florida, USA (Russel’s Bromeliads) before being housed in the Colgate University greenhouse (86% humidity and 27° C). Before experimental treatment, each leaf length was measured and labeled (new leaves were defined as leaves that emerged after this initial labeling and measuring period). *V. cathy* is a tank bromeliad, which means that its leaves develop around a “tank” like container in the middle where water collects. Nutrients from either wet or dry deposition into the tank are absorbed through the leaves surrounding the tank. Leaves grow externally from the inside of tank. The tank morphology allows *V. cathy* to retain moisture for extended periods of time (Zotz & Laube 2005), which Martin (1994) identifies as critical towards maintaining stomatal conductance and photosynthetic rates in temperature and water stressed environments. Additionally, the size of the plant plays a disproportionate role in water retention and must be controlled for (Zot & Thomas 1999). *T. bulbosa* is an atmospheric bromeliad (lacking roots and an overlapping leaf base) that absorbs nutrients and water from specialized foliar trichomes in their shoots (Benzing & Dahle 1971). Though this allows *T. bulbosa* to resist desiccation in temperature and water stressed environments, juvenile *T. bulbosas* are more susceptible to temperature and water stress and therefore its tolerance is not consistent across ontogeny.

**Experimental Treatments and Design**

I had two treatment temperatures (27° C and 32° C) and four nutrient levels within each treatment (N, P, NP, and a control [C]). Humidity within each growth chamber was provided by continuous water vapor input via a portable humidifier. In addition to the humidifier, the plants were watered every three days. Eighty individual plants from each species were used in total. Forty individuals of each species were placed in either an ambient or a 5° C above ambient growth chamber.
respectively. Within each temperature treatment, ten individuals were randomly assigned each nutrient level.

The C treatment broth was made up of Bold’s Medium and each successive treatment consisted of the Bold’s Medium with a doubling of the respective nutrient (e.g. double nitrogen for the nitrogen treatment). Fifteen milliliters (mL) of each nutrient broth was added to each individual plant; however, due to the physiology of each plant, the solutions were applied in different ways. 15mL of nutrient media was poured directly in Vriesia cathyi tanks, while 15mL of nutrient media were sprayed onto T. bulbosa.

I marked and measured new leaf growth as growth proceeded. New leaves were monitored on three separate occasions for T.bulbosa (Tbt1-t3) and two times for V. cathyi (Vct1-t2), the difference in time between measurement was taken into account when calculating growth rates. Similar to using RWR as a proxy for nutrient acquisition, leaf length can provide an estimate for response to nutrient levels because epiphytes uptake nutrients through their leaves.

**Statistical Analysis**

Relative growth rates were calculated by re-measuring these new leaves and determining their percent growth as a function of time. Growth rate was calculated as the difference between t3-t1. The number of clonally produced buds were also noted for each individual and each leaf on the bud was marked. Biomass was determined by measuring new leaf length. Percent biomass growth was measured by adding up the total new leaf length of a plant and using that value to calculate new leaf growth as a percentage of initial total leaf growth in order to control for initial plant size. The number of new leaves produced was calculated as a function of initial biomass. The statistical effect of fertilization on leaf length, growth rates, biomass, and bud production was determined using an ANOVA. Across species tests only compared initial leaf size and rate of growth which controlled for days that the plants grew in between sampling periods. Letters are used to denote significance within species whereas numbers are used to denote significance across species.

**Results**

1) **Analysis of Growth as a Function of Temperature Treatment**

*Tilandsia bulbosa*

New leaf growth was compared over time and between temperature and
nutrient treatments. The data were not normally distributed (Test for Normality, \( p = 0.001 \)) and so they were square-root arcsine transformed. Plants that did not grow any new leaves were assigned a value of zero. There was a significant effect of nutrient treatment on plant growth (Figure 1, \( F_3 = 3.626, p = 0.018 \)).

There was a significant effect of temperature on plant growth across treatments (Oneway ANOVA, \( F = 7.46, p = 0.0081 \)). Within the P treatment, the plants in the 32° chamber grew significantly less than the plants in the 27° chamber (Figure 1, Wilcoxon/Kruskal-Wallis Tests, \( S = 33, Z = -2.896, p = 0.0038 \)). There was no significant effect of temperature (MANOVA \( F_1 = 0.976, p = 0.327 \)) on new leaves produced.

Vriesia cathyi

Biomass and growth rates were, on average, significantly higher in the 27° treatment. Temperature had a significant effect on the biomass production in sampling period Vct2 (ANOVA \( F_1 = 4.599, p = 0.036 \)) and on the growth rate of plants across the whole sampling period (ANOVA \( F_1 = 4.202, p = 0.044 \)).

2) Effects of Nutrient Treatment on Growth

Tilandisa Bulbosa
T. bulbosa Individuals in the 27° treatment grew significantly more under N and P separately than with NP combined (Student’s t-test, $t = 2.0289$, $p = 0.0027$). There was no significant effect of nutrients on new leaf growth rates in the 32° treatment (One way ANOVA, $F_{3} = 1.59$, $p = 0.22$). There was no significant effect of nutrient treatment (MANOVA $F_{3} = 2.059$, $p = 0.114$) on number of new leaves produced. There was also no significant effect nutrient treatment (MANOVA $F_{3} = 0.512$, $p = 0.675$) on growth rate of new leaves (See figures 1 & 2). Under ambient conditions, P-fertilization had the predicted effect of increased growth; however, under higher temperatures this was not the case (insert figure). The latter response is likely due to increased stress associated with higher temperatures.

Vriesia cathyi

V. cathyi saw a marginal significant effect of nutrient treatment on relative biomass ($F_{3} = 2.55$, $p = 0.073$); within nutrients but across temperatures, the N treatment was the only one with marginal significance ($F_{1} = 3.77$, $p = 0.072$). Because these results are only from 30 days of growth, these data suggest that there is some significant interaction of nutrient treatment on growth.

3) Effects of Development on Growth and Across Species

There was no significant effect of temperature (MANOVA $F_{1} = 2.835$, $p = 0.097$) on growth rate of new leaves in T. bulbosa; however, new leaves of V. cathyi grew significantly slower in the 32° treatment (as shown above). Because there was no significant effect of nutrient treatment on growth rate of new leaves within species, all the nutrient categories were combined for the across species comparison.

There was no significant difference in biomass of initial leaves produced under treatment (ANOVA $F_{1} =$...
-0.007, p = 0.933); however there was a significant effect of species on growth rate (ANOVA F1 = 53.8, p = 0.000). Within the 27° treatment, V. cathyi leaves grew significantly faster in the N treatment (F1 = 15.5, p = 0.001), NP treatment (F1 = 38.2, p = 0.000) the P treatment (F1 = 5.06, p = 0.037), and the C treatment (F1 = 4.85, p = 0.044). Within the 32° treatment, V. cathyi leaves grew significantly faster in the N treatment (F1 = 10.43, p = 0.007) and the NP treatment, (F1 = 15.70, p = 0.001) but not the P treatment (F1 = 2.66, p = 0.122). New leaves of Vriesia cathyi grew significantly faster than new leaves of Tilandsia bulbosa.

Discussion

Combined Factors of Nutrients and Temperature

This study provides a first glance at the effects that increased temperature and different nutrient availabilities have on epiphytic bromeliad plant growth. Within treatment variance was higher in the 27° dataset than the 32° dataset. This suggests that there is some selective pressure that is restricting the variation in plant phenotypic response concerning leaf growth at the higher temperature. The restricted variation within the 32° dataset may be one of the reasons that the lack of an effect of nutrient treatment on plant growth in that environment. Various ecosystems around the globe see differential nutrient availability (Cornwell & Grubb 2003) and consequently, previous studies hypothesized that global warming would effect different ecosystems with respective nutrient availabilities differently (Vitousek 1994). Especially because previous studies hypothesized that increased nutrient availability might decrease species biodiversity (Bedford et al. 1999), our study showing that there might not be a difference in response to global warming across habitats with differing nutrient availabilities and that the effects of nutrient availability diminish in a temperature stressed environment are quite relevant. These results are similar to those found in other studies (King et al. 1999).

Phosphorus has been hypothesized to be the limiting nutrient in epiphytic communities (Zotz & Richter 2006), which may be one reason why we saw a significant effect of temperature on the P treatment plants. Temperature’s effect on growth is intensified when looking at limiting nutrients; consequently, the 27° P fertilized plants were able to grow significantly more than the 32° P fertilized plants. Because the 27° plants had excess of their hypothesized limiting nutrient, (P), their growth was only inhibited by factors such as water availability but not temperature stress. In the 32° treatment, the P fertilized plants may have had excess of their limiting nutrient as well, but they also had the increased stress of higher temperatures. Even having an excess of P in the 32° chamber was not enough to counteract the stresses of having a higher environmental temperature.

Nutrient immobilization might be the reason why the NP fertilized plants showed significantly less new leaf growth than the P and the N fertilized plants in
the 27°C treatment. The plants could have been responding to the surplus in nutrients by immobilizing them in order for them to be available for future growth. Luxury consumption has been shown to be present in P fertilization studies on bromeliads (Winkler & Zotz 2008). Another factor may be that the stress of increased temperature reduces water use efficiency (due to elevated ET) and therefore the plants need to keep their stomata closed more and are subsequently not able to uptake enough nutrients to display significant differential growth with respect to nutrient availability. It was hypothesized that variation in nutrient availability results in variation in leaf structure and plant response to environmental stressors (reviewed by Gutschick (1999)) which we say in terms of growth rate and new biomass produced; however, data from future analyses is necessary in order to fully explain the effects that different nutrient loads are having on plant growth.

**Temperature**

That temperature showed a significant effect in both the Vriesia cathyi and Tillandsia bulbosa suggests that temperature was an important factor in these epiphytic communities for governing growth. Interestingly, in the bulbosas, temperature did not show a significant effect on the growth rate of newly produced leaves but it did show an effect on percent relative leaf growth (for the phosphorus treatment). Evolutionarily, this might mean that after plants produce new leaves that develop in the changed habitat (32°C chamber), they are able to phenotypically adapt through their development in the higher temperature and subsequently grow at similar rates to those produced in the original habitat (27°C chamber). While the original, fully developed plant, decreases its total amount of growth in a warmer temperature, the new leaves that develop in the warmer temperature do not. This could mean that development plays a certain role in governing plasticity in temperature tolerance. By this, I mean that instead of phenotypic plasticity being constant throughout ontogeny, it may be more sensitive or at least more able to change during development of new physiology. Contradicting my bulbosa data are the cathyi data that show a significant effect of temperature on growth rate of newly produced leaves, with the 32°C leaves growing more slowly. A possible explanation for the difference across species could be due to how the leaves develop in their respective plant types.

The emergence of new leaves in the bulbosa species was more immediate because they emerge from already existing elongated leaves, with whole leaves sometimes emerging fully grown. The bulbosa leaves seem to grow inside of already established leaves and then emerge for unknown reasons. Cathyi leaves grown from the middle tank and are external for their entire measured length. These physiological traits can be presented by the data that showed the cathyi having a significantly greater growth rate of new leaves than bulbosa; therefore, the cathyi leaves are developing externally significantly more so than the bulbosa leaves. Therefore, there are unexplained physiological
characteristics that seem to be at play that might be able to explain some of these differences. Another potential reason why there was no difference in growth rate between treatments is because the T. bulbosa leaves do not grow very much after they emerge and subsequently there is not enough measurable new leaf growth within the time frame of this study. In order to draw concrete conclusions as to what is really going on during leaf development, a more in depth study of leaf development comparing across species is necessary to explain the difference in response.

This study has laid the groundwork for future studies that can perform more in depth analyses of factors controlling growth in the plants. A more in depth study using nutrient isotopes will be useful in showing us where physiologically the plants are placing the nutrients. A study keeping track of individuals and their clonal offspring is necessary also in order to tease apart how different stages of development affect phenotypic plasticity.

Bibliography


IPCC (Intergovernmental Panel on Climate Change), Climate Change 2007: The Physical Science Basis, Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change (Solomon, S., D. Qin, M. Manning, Z. Chen, M.


