Predator phylogenetic diversity decreases predation rate via antagonistic interactions

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## Introduction

Predators can have strong top-down effects, both on community structure and ecosystem processes (Estes et al. 2011); however the effects of predator species together (i.e., of predator species richness) are not well understood. The effect of predator species richness on communities is often stronger or weaker than what might be predicted from a study of those same species in isolation (Sih, Englund, and Wooster 1998; Ives, Cardinale, and Snyder 2005). These non-additive effects of predator richness occur when predators interact directly with each other, or indirectly via prey species. For example, predators feed directly on each other (intra-guild predation), consume the same prey (resoon-adurce competition) or modify the behaviour of other predator species or prey via nonconsumptive (i.e. trait- mediated) interactions (Sih, Englund, and Wooster 1998; Griswold and Lounibos 2006; Nyström et al. 2001). These non-additive effects can be positive or negative: e.g. prey may have an induced a defense against one predator which makes it harder (negative non-additive effect) or easier (positive non-additive effect) for another predator to eat it. While there are many possible mechanisms underlying the effect of predator composition, we lack a means of predicting *a priori* the strenght and direction of predator diversity on communities and ecosystem functions.

Predator phylogenetic diversity could predict the effect of predator richness on ecological communities. Measures of phylogenetic diversity include the evolutionary relationships of species into measures of diversity. The measurement of phylogenetic diversity has become a popular means for ecologists to make inferences about ecological and evolutionary mechanisms behind observed patterns in natural communities (Cavender-Bares et al. 2009). For example, the phylogenetic diversity of plant communities is a better predictor of productivity than either species richness or diversity (e.g. M. W. Cadotte et al. 2009; M. W. Cadotte, Cardinale, and Oakley 2008; Godoy, Kraft, and Levine 2014). A central assumption of this approach is that increased phylogenetic distance implies increased ecological dissimilarity -- either in the form of differences in species niches, interactions, or functional traits. When this is true, high phylogenetic diversity leads to complementarity between species, resulting in increased ecosystem functioning (D. S. Srivastava et al. 2012). Despite the prevalence of phylogenetic community ecology and the importance of predators to natural systems, the phylogenetic diversity of local predator assemblages has rarely been measured (L. Bersier and Kehrli 2008; Naisbit et al. 2011). Many studies of phylogeny and predator traits focus on whole clades, rather than local assemblages (e.g. *Anolis* lizards (Knouft et al. 2006), warblers (**???**- Gaese2003), tree boas (Henderson, Pauers, and Colston 2013) and wasps (Budriene and Budrys 2004)) making it difficult to connect these results to predator effects at the scale of a local community. These clade-specific studies often find weak evidence for phylogenetic signal in ecologically- relevant traits; in contrast, studies at the level of the whole biosphere (Gómez, Verdú, and Perfectti 2010; L. Bersier and Kehrli 2008) demonstrate that related organisms often have similar interspecific interactions -- i.e. related predators often consume similar prey. In order to understand how predator diversity will affect community composition and ecosystem function at the local scale, we need to understand both their distribution and their interspecific interactions alone and in combination.

Within a local community, the effect of predator species diversity will depend on three factors: how predators are distributed among habitats, how they interact with prey, and how they interact with each other. To the extent that phylogenetic relationships are correlated with all of these factors, they can predict the impact of predator diversity on communities. For instance, phylogeny could constrain predator species composition if more distant phylogenetic relatives have more distinct fundamental niches, while close relatives are too similar to co-exist (Webb et al. 2002; Emerson and Gillespie 2008). When predators do co-occur, phylogeny may correlate with their feeding behavior, such that more closely related predators consume more similar prey. Diet overlap (shared prey species between predators) will depend on the feeding traits and nutritional requirements of predators -- both of which may be phylogenetically conserved. If this is the case, then predator assemblages with higher phylogenetic diversity will show greater prey consumption and therefore a stronger top- down effect on ecosystem function (Finke and Snyder 2008). In some cases, predator diets may extend to include other predators, leading to direct negative interactions such as intraguild predation, which may also have a phylogenetic signal (Pfennig 2000).

We examined the predator community using a natural mesocosm: the community of macroinvertebrates living within bromeliads. Bromeliads (Bromeliaceae) are flowering plants native to the Neotropics. Many bromeliad species contain water and detritus; the decomposition of this detritus supplies nutrients for the bromeliad (**???**). The small size of these habitats permits direct manipulations of entire food webs, manipulations which would be difficult in most natural systems. Within this aquatic food web, damselfly larvae (e.g. *Leptagrion* spp., Odonata:Coenagrionidae) are important predators; the predator dramatically alters community dynamics (e.g. decreases insect emergence (Starzomski, Suen, and Srivastava 2010) and increases nutrient cycling (Ngai and Srivastava 2006)). However, other predators are also found in bromeliads, including large predaceous fly larvae (Diptera: Tabanidae) and predatory leeches.

We used a series of observations, lab feeding trials, and manipulative field experiments to measure how the phylogenetic diversity of the predator assemblage predicts community composition and ecosystem function. We test three related hypotheses concerning similarity in distribution, diet and top- down ecosystem effects of predators,

1. *Distributional similarity*: closely-related predators may occur together more frequently than less-related predators if there is a strong phylogenetic signal to habitat requirements. Alternatively, very closely related species may never co-occur because high overlap in ecological niches results in competitive exclusion.
2. *Diet similarity*: similarity in diet (as measured by feeding trials) decreases with phylogenetic distance if diet is conserved. Alternatively, closely related species may have evolved different diets to allow coexistence.
3. *Ecosystem-level effects*: our experiments at the level of the whole habitat patch (i.e. a single bromeliad) allow us to test hypotheses about direct and indirect effects of predator combinations:
   1. Closely-related predators will have similar effects on the community. This will occur if related predators share similar trophic interactions (e.g. predation rate, diet similarity). Our monoculture treatments allow us to assess the effect of each predator both on prey survival and on ecosystem function (Nitrogen uptake and decomposition).
   2. Predator assemblages with higher phylogenetic diversity will show a stronger non-additive effect. This will occur if phylogenetic distance correlates with increasing trait difference, and if this trait difference in turn results in niche complementarity. At the extreme, different predators may consume each other. By comparing treatments with pairs of predators to treatments that received a monoculture of each predator, we are able to estimate additive and non-additive effects.

## Methods

## Study Design

We conducted this work in Parque Estadual da Ilha do Cardoso in São Paulo state, Brazil, in 2011. We collected three datasets to address each of these questions. We examined distributional similarity among predator species (Question 1) by making observations of predator distribution among habitat patches. We examined diet similarity (Question 2) with a series of no-choice feeding trials in under laboratory conditions. Finally, we examined the effects on predators on whole communities (Question 3) with a field experiment, in which predators were added to habitat patches containing standardized communities of prey. This experiment included both single species treatments and two species treatments; the latter were chosen to create the widest possible range of phylogenetic diversity.

In each dataset, we related the results to published phylogenies accessed from "timetree.org", an online database of published molecular time estimates (Hedges, Dudley, and Kumar 2006). Node age data was available for all but the youngest nodes, where either a lack of taxonomic information (e.g. Tabanidae) or a lack of phylogenetic study (e.g. *Leptagrion*) prevented more information from being included. These branches were left unresolved and were all assigned identical, arbitrary and short branch lengths (15 Mya).

We conducted all three experiments in Parque Estadual da Ilha do Cardoso ( S, W), a 22.5 ha island off the south coast of São Paulo state, Brazil. We worked in a coastal forest (*restinga*) in the understory of which grows the abundant *Quesnelia arvensis* Mez. (Bromeliaceae). *Q. arvensis* is a large terrestrial bromeliad that catches and holds rainwater (phytotelmata), accumulating up to 2.8 L of rainwater in a single plant. Our observational survey found more than 47 species of macroinvertebrates in these aquatic communities (Romero and Srivastava 2010), in ncol(metabolic) -1 bromeliads of various sizes. This diversity encompasses multiple trophic and functional groups: filter feeders (Diptera:Culicidae); detritivores including shredders (Diptera:Tipulidae, Trichoptera:Calamoceratidae), scrapers (Coleoptera:Scirtidae), and collectors (All Diptera:Chironomidae, Syrphidae, Psychodidae). These species are the main prey for a diverse predator assemblage of at least three species of damselfly larvae (*Leptagrion* spp., Odonata:Coenagrionidae), two species of predatory Horse Fly larvae (Diptera:Tabanidae), and two species of leech (Arhynchobdellida).

## Data collection

### Question 1: Distributional similarity

To examine distributional similarity among predator species, we used a detailed survey of bromeliad communities. In 2008, each bromeliad was dissected and washed to remove invertebrates and the resulting water was filtered through two sieves (250 and 850 µm), which removed particulate organic matter without losing any invertebrates. All invertebrates were counted and identified to the lowest taxonomic level possible. The body length of all individuals was measured when possible for small and medium-sized taxa (< 1cm final instar) and always for large-bodied taxa (> 1 cm final instar).

### Question 2: Diet Similarity

To examine diet similarity among predator species, we fed prey species to predators in laboratory feeding trials. We conducted 314 feeding trials of 10 predator taxa and 14 prey taxa between March and April 2011. We covered all potential predator-prey pairs present in the experiment (described below), and attempted to perform all other trials whenever possible. However, due to the rarity of some taxa, many predator-prey pairs were not possible; we tested 56 pairwise combinations. Most trials were replicated at least five times, but the number of replicates ranged from 1 to 11. To conduct the trials, we placed predators together with prey in a 50ml vial, with a stick for substrate. The only exception was the tabanid larvae, which we placed between two vertical surfaces to imitate the narrow space found in bromeliad leaf axils (their preferred microhabitat; necessary for successful feeding). Generally our trials contained a single predator and a single prey individual, except in the case of very small prey (*Elpidium* sp.) or predators (*Monopelopia* sp.) in which case we increased the density. We recorded whether prey was consumed after 24 hours.

### Question 3: Community effect experiment

Our third question had two parts: (a) how do predator species differ in their effects on the whole community and (b) does predator diversity show non-additive effects on the community, and do these non-additive effects increase or decrease with phylogenetic distance? We measured five community and ecosystem response variables: production of fine particulate organic matter (FPOM), decomposition of coarse detritus, bromeliad growth, uptake of detrital nitrogen into bromeliad tissue, and survival of invertebrate prey (emerged adults + surviving larvae)

#### Experimental design

In this experiment we focused on the four most abundant large predators found in this community: *Leptagrion andromache* and *Leptagrion elongatum* (Odonata: Coenagrionidae), a predatory Tabanid fly (Diptera:Tabanidae:*Stibasoma* **correct?**) and a predatory leech. We combined these species in eight treatments: predator-free control (no predators), each of the four predator species alone (part a) and pairs of predator species chosen to maximize the range of phylogenetic distance (part b). Specifically, these pairs were: two congeneric damselflies (*Leptagrion andromache* and *Leptagrion elongatum*), two insects (*L. elongatum* and a Tabanid predatory fly), and two invertebrates (*L. elongatum* and a predatory leech). We used five replicate bromeliads for each treatment (8 treatments, n=5). In order to control for differences in body size and feeding rate among predator species, we used a substitutive design which maintained the same predator metabolic capacity in all replicates (see below). Substitutive experiments often hold total abundance constant, but when species differ substantially in body size - as in this experiment - allometric effects of body size on feeding rate can confound detection of effects based on trophic traits or species interactions, and standardizing to community metabolic capacity is preferred (D. S. Srivastava and Bell 2009). This experiment allows the estimation of the effect of each predator species (monoculture treatments), as well as the detection of non-additive effects in predator combinations.

We used an allometric scaling relationship to standardize "metabolic capacity" of predators across treatments. Species co-occurrence is often measured in terms of non-random patterns of species presence/absence or abundance, but such measures will only be poorly related to the functional effects of species when species differ substantially in body size. Integrating the allometric relationship between body size and feeding rate (Brown et al. 2004; Wilby, Villareal, and Lan 2005) over all individuals of a species allows estimates of "metabolic capacity", or the potential energy requirements of a species (D. S. Srivastava and Bell 2009). Metabolic capacity is equal to individual body mass raised to the power of 0.69 (an invertebrate-specific exponent determined by Peters 19xx and confirmed by Chown et al, (2007)); this reflects the nonlinearity of feeding rate on body size across many invertebrate taxa. We used metabolic capacity to inform both our observational results and our experimental design (details below), with the exception of our feeding trial data. This is because the feeding trials were intended to measure which prey species our predators ate, rather than their feeding rate (only the latter should scale with metabolic capacity).

In Feburary 2011 we collected bromeliads with a volume between 90 and 200ml, thoroughly washed them to remove organisms and detritus, and soaked them for 12 hours in a tub of water. We then hung all bromeliads for 48 hours to dry. This procedure was indended to remove all existing insects; one bromeliad dissected afterwards contained no insects. Each bromeliad was supplied with dried leaves of the species *Plinia cauliflora*, simulating natural detritus inputs from the canopy. In order to track the effects of detrital decomposition on bromeliad nutrition, we enriched these leaves with 15N by fertilizing five *Plinia cauliflora* (Jabuticaba, Myrtaceae; a common Brazilian tree) plants with 40ml pot-1 day-1 of 5g L-1 ammonium sulphate containing 10% atom excess of 15N over 21 days. Whole leaves were then picked from plants, air-dried until constant weight, then soaked for three days and the water discarded. This procedure removes excess nutrients from the artificial fertilization. About 1.5 g of leaves were added to each bromeliad (1.5g ± 0.02).

Each bromeliad was stocked with a representative insect community. The densities of each prey taxon were calculated from the 2008 observational dataset, using data from bromeliads of similar size to those in our experiment. All densities used were within the range of these calculated abundances, and all experimental bromeliads received the same insect community. Halfway through the experiment, insects were added to bromeliads a second time to simulate the continuous oviposition that characterizes the system. Throughout the experiment, all bromeliads were enclosed with a mesh cage topped with a malaise trap and checked daily for emergence of adults. At the end of the experiment we completely dissected our bromeliads, collecting all invertebrates and detritus remaining inside.

To quantify the effect of predators on ecosystem function, we measured five community and ecosystem response variables: production of fine particulate organic matter (FPOM), decomposition of coarse detritus, bromeliad growth, uptake of detrital nitrogen into bromeliad tissue, and survival of invertebrate prey (emerged adults + surviving larvae). We measured the decrease in coarse detritus, and the increase in fine, by separating the collected detritus on a sieve and air drying it to constant weight before weighing. We measured uptake of labelled detrital nitrogen by analysing three samples of bromeliad leaves. Finally, we quantified the species composition and survivorship of invertebrate prey by adding together counts of emerging adult insects with counts of surviving larvae.

## Data analysis

### Question 1 and 2: similarity in distribution and diet

We quantified the effect of phylogenetic distance on each of distributional (Question 1) and diet (Question 2) similarity. First, we calculated phylogenetic distance between each pair of species, then fit several functions to the relationship between phylogenetic diversity an either distributional or diet similarity. We used linear, constant, and several appropriate nonlinear functions (nonlinear, because our measures of similarity are bounded by 0 and 1; see below). We compared these models using AIC and generated confidence intervals as appropriate (parametric or bootstrap for linear and nonlinear, respectively). We evaluated both distributional and diet similarity between predators using Pianka's index of niche overlap (Pianka 1974):

For each pair of predators, and represent the preference of predator or for resource or habitat . The value represents similarity (in our case, in either distribution or diet) and ranges from 0 (complete dissimilarity) to 1 (complete similarity). The resources represent the different habitats surveyed for Question 1 (distributional similarity), or the different prey species assayed for Question 2 (diet similarity). Preference () represents the proportion of a predator's total metabolic capacity found in a particular bromeliad (Question 1); or the proportion of feeding trials in which it ate a particular prey (Question 2).

### Question 3: similarity in community effect

We tested effects of both single or multiple predator species on community responses with a manipulative experiment where identical communities were exposed to treatments of either a single predator, or pairs of predators representing increasing phylogenetic diversity. We divided the analysis of this experiment into three parts, quantifying the effect of phylogenetic distance on prey community similarity, community responses, and non-additive effect. First, we compared the four treatments with single predator species by calculating the similarity in species composition (Pianka's index) between surviving prey communities and relating this to the phylogenetic distance between predators. If predator feeding choices are phylogenetically conserved, that diet similarity will decline with increasing phylogenetic distance.

Second, we examined how predators affected the five community and ecosystem responses we described above, testing in turn the effect of predator presence, number, species identity, and finally phylogenetic diversity. For each response, we combined our treatment levels into four different contrasts, each of which examine a different aspect of predator impact. To test for an effect of predator presence, we contrasted the control treatment (predators absent) with the mean responses of all seven treatments that did contain predators. To test for an effect of predator species number (one or two predators), we contrasted the means of all monocultures with the means of all pair treatments. We compared all four single-species treatments to test for an effect of predator identity, and finally we tested for an effect of increasing phylogenetic diversity among the three two-species treatments. We analyzed each of these responses with one-way ANOVA.

In our third and final analysis we quantified the nondditive effect of predator species. We calculated this effect as the difference between the response in bromeliads with both predator species (n=5) and the mean response in bromeliads with either one of these two predator species (n=5 for each predator species). We generated bootstrap confidence intervals for these non-additive effects; confidence intervals which do not overlap zero indicate a significant non-additive effect of a predator combination.

## Results

### Question 1 and 2: similarity in distribution and diet

We did not find any significant relationship between habitat distribution (measured as Pianka's index of niche overlap) among predator species and the phylogenetic distance between them (Figure 1a, F1,89=2.39, p=0.13). Indeed we often found multiple predator species co-occurring in the same bromeliads (mean 4.4 ± 2.9 predator species per plant). This indicates that predator species all have roughly similar habitat distributions at the level of the bromeliad. We found this pattern of broad overlap despite this dataset containing more more species than our other datasets, a total of 14 predator taxa in 25 bromeliads. We were able to sample a wide range of phylogenetic relatedness, including two groups of congenerics -- two species of *Bezzia* sp. (Diptera:Ceratopogonidae) and three species of *Leptagrion* sp. (Odonata:Coenagrionidae). There were also two groups of confamilials -- three species of Tabanidae and two species of Empididae, all Diptera. Deeper divisions were also present: three families of Diptera are represented by a single predator species each (Dolichopodidae, Corethrellidae and Chironomidae) and the deepest taxonomic divide is between all insects present and the predatory leeches (Annelida:Arhynchobdellida).

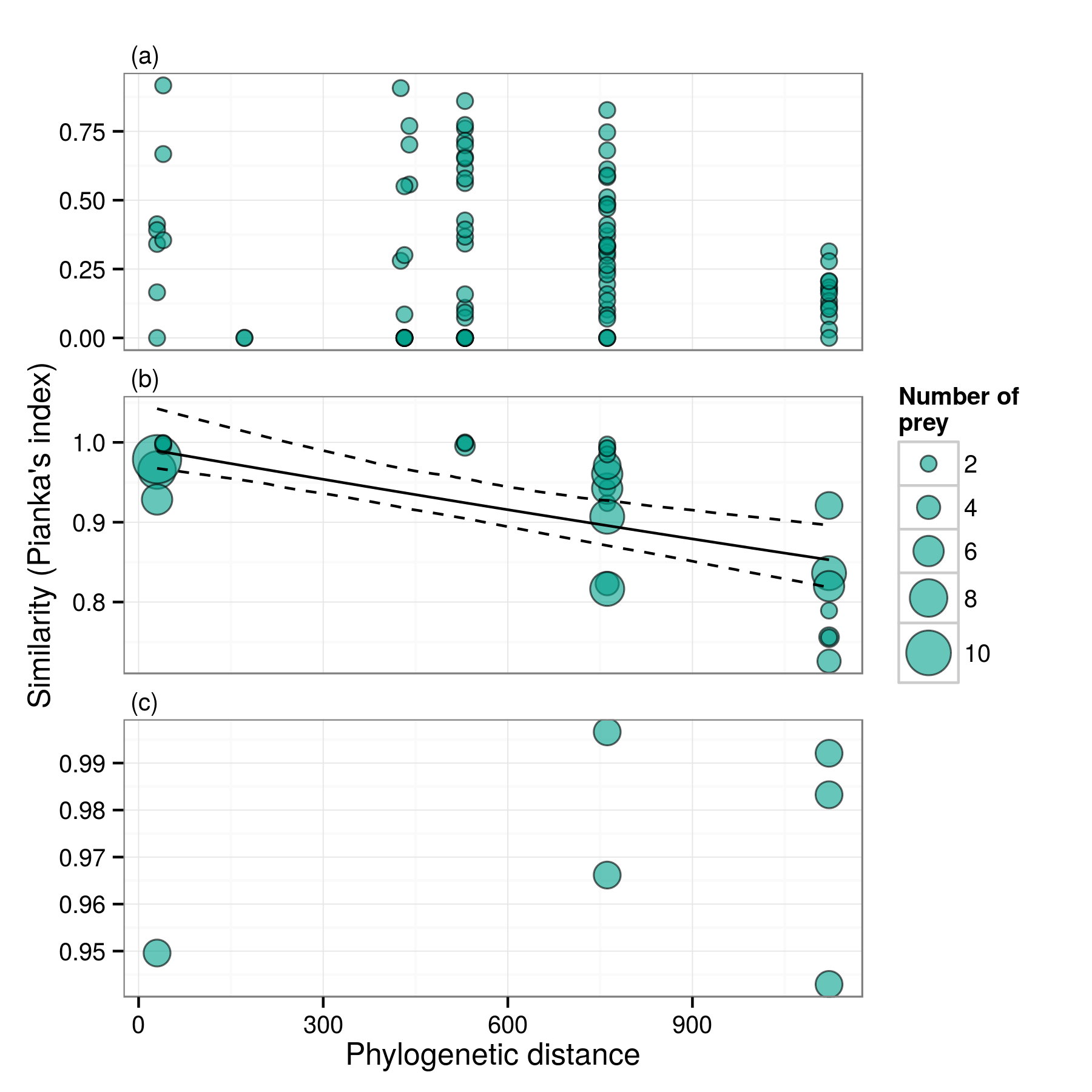
More phylogenetically distant predators differed in their preference of prey species, as measured by the niche overlap index (Fig 1b, F1,26=19.41, p=0.00016), regression weighted by the number of prey species assayed.). Among the most common predator taxa (i.e. those used in our experiment, described below) the damselflies (*Leptagrion andromache* and *Leptagrion elongatum*) showed the highest rates of prey consumption (prey consumed in 94.3% and 67.7% of trials, respectively). Despite the decline in diet Similarityy with phylogenetic distance, the variation in predator feeding behaviour did not translate into a significant difference in the composition of prey species surviving the manipulative experiment (Fig 1c, F1,4=0.6, p=0.48)

### Question 3: similarity in community effect

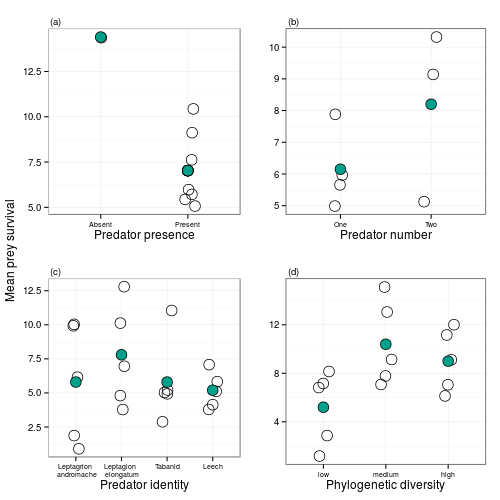
We analyzed our five response variables from the manipulative experiment by dividing them into four separate tests: predator presence, predator number, predator species identity and, increasing predator phylogenetic diversity. Across all four tests, we saw the strongest responses for total prey survivorship (Table 1). Survivorship was reduced by nearly half when predators were present (Figure 2a, Table 1). Survivorship was reduced by the same amount by all predators (Figure 2c, Table 1); however increasing predator phylogenetic diversity caused a significant increase in total prey survivorship (Fig 2d). That is, more phylogenetically diverse predator combinations caused less mortality for prey. Interestingly, these effects on prey density did not result in a change in the processing of detritus (reduction in coarse detritus or production of fine detritus), bromeliad growth or nitrogen cycling (Table 1).

We tested for non-additive effects of phylogenetic diversity with a randomization test, and once again found the greatest effect for prey survival. While effects of *L. andromache* and *L. elongatum* in combination were quite similar to the effect of either alone, when *L. elongatum* was placed in the same plant as either a Tabanid larva or leeches, on average five more prey individuals (18% of total prey community) survived till the end of the experiment (Fig 3). Once again, this effect on invertebrate density did not in turn create a significant difference in other response variables.

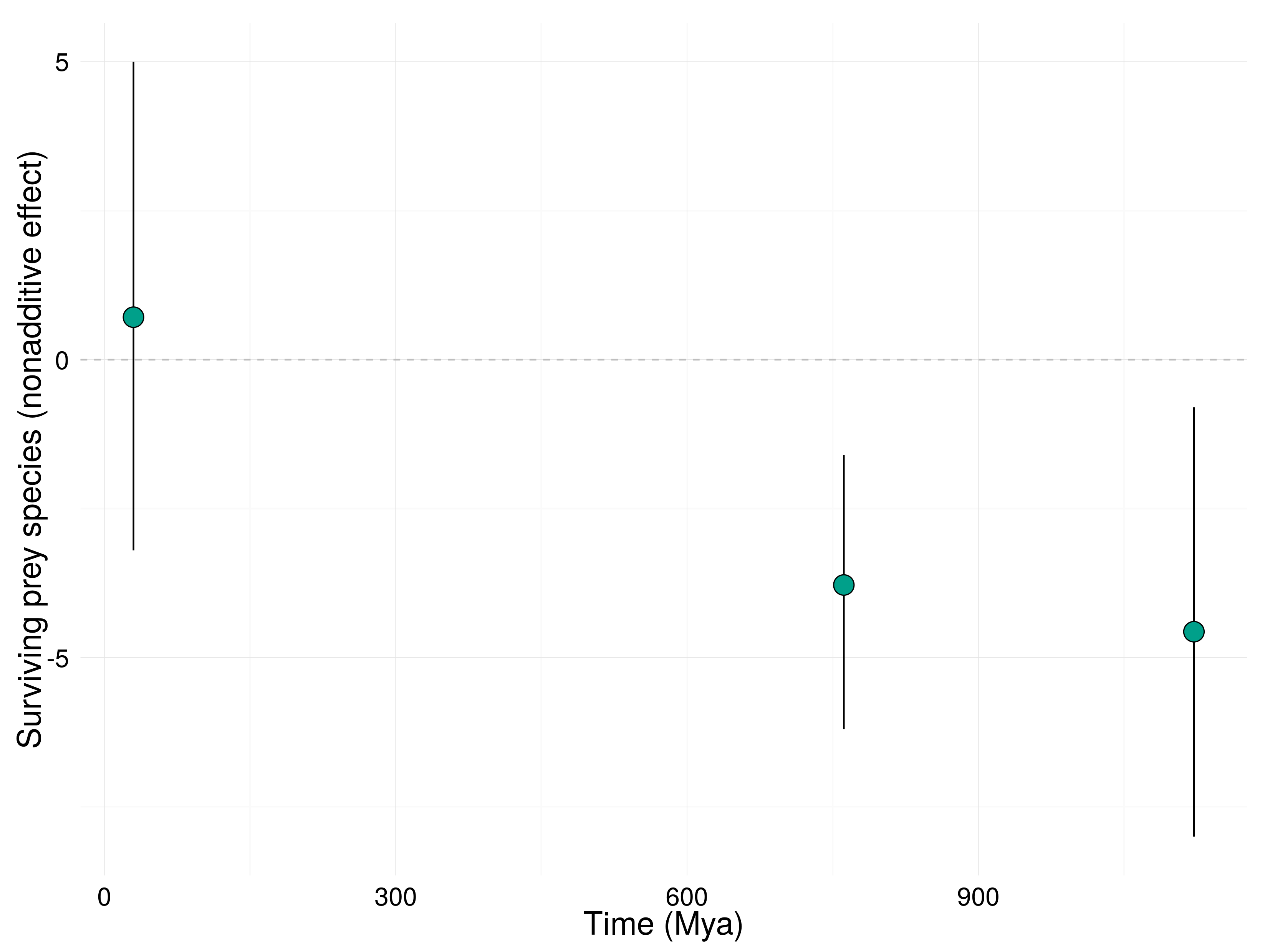
### Figures



**Figure 1**: Phylogenetic distance and niche overlap among predators. Our measures of niche overlap were: (a) distribution among bromeliads; (b) diet preferences and (c) community composition of surviving prey. We measured distributional similarity (a) by counting all predators in 25 bromeliads, estimating their total metabolic capacity, and calculating niche overlap among all pairs of species. We measured diet preferences (b) for a subset of these predators by offering them various prey in no-choice trials. Finally, we measured community composition of surviving prey (c) at the end of an experiment in which predators were placed in bromeliads with standardized communities (see main text for details). We used Pianka's index of niche overlap and fit various nonlinear models (see Appendix) to the relationship between this index and phylogenetic distance. Solid lines show signifigant model fit, and dashed lines show bootstrap 95% quantiles.



**Figure 2**: The effect of predators on the survival of prey organisms. We show the effects of predator presence (a), increased number of predators (b), predator species identity (c) and predator species pairs (d, arranged in order of increasing phylogenetic distance). Shaded dots represent grand means for each group; unshaded dots are either treatment means (2a and 2b, n = 5) or individual bromeliads (2c and 2d).



**Figure 3**: Phylogenetic distance and non-additive effects of predator combinations. We calculated non-additive effect size by first subtracting treatment means from control (no predators), then subtracting the mean of single-predator treatments from two-predator treatments. A difference of 0 indicates that two-predator treatments resulted in no more prey mortality than would be expected from simply averaging single-predator treatments. Error bars represent bootstrap 95% confidence intervals.

**Table 1** Predator diversity effects on community and ecosystem variables. We measured four community-level variables: total prey survival (both emerged adults and surviving larvae), the breakdown of coarse detritus, the production of fine particulate organic matter (FPOM), and the growth of the bromeliad itself. We contrast treatments in our experimental design in three ways: comparing treatments with predators to those without, contrasting predator species, comparing predator communities of 1 or 2 species, and considering the effects of phylogenetic distance between predators. \* = p < 0.05, \*\* = p < 0.01

Response | Predator Presence | Identity | Richness | Pairwise PD |  
-------- | ------------------| ---------|--------- | ---------- |  
Total prey survival | F1,10 = 9.07\* | F3,16 = 0.6 | F1,5 = 1.96 | F1,13 = 7.64\* |  
Decomposition (g) | F1,10 = 0.47 | F3,15 = 1.29| F1,5 = 0.21 | F1,13 = 0.4 |  
FPOM (g) | F1,10 = 0.92 | F3,16 = 0.42 | F1,5 = 6.47 | F1,13 = 1.35 |  
Bromeliad growth | F1,10 = 0.51 |F3,16 = 0.96 | F1,5 = 0.49 | F1,12 = 1.29 |  
Nitrogen cycling | F1,10 = 2 |F3,16 = 1.84 | F1,5 = 0.5 | F1,13 = 0.15 |

## Discussion

We found that in our bromeliad system, phylogenetically distant pairs of predators are distributed with the same degree of similarity as phylogenetically similar predators (Question 1). However, we did find that phylogenetically distant predators had slightly different diet preferences (Question 2). Interestingly, these apparent diet preferences in the lab did not create a difference in surviving species composition in the field (3a). Phylogenetic diversity caused an increase in prey mortality (i.e. a decrease in predation); phylogenetically diverse predators pairs also showed a significant negative non-additive effect (3b).

### Phylogenetic distance and similarity in distribution?

We found no relationship between phylogenetic distance between predator species and similarity in their distribution among habitats. This similarity in distribution could be caused by two processes: low habitat variability among bromeliads, or low preference of predators for different habitats. We already know that bromeliads vary widely in abiotic conditions, size, detritus amount and prey community, therefore it seems unlikely that low patch variation explains the lack of pattern. It appears instead that predators do not possess any strong phylogenectically-conserved preferences for different habitat characteristics, showing instead very generalist habitat preferences. This is not uncommon among organisms which live in small, fluctuation-prone habitats (). Additionally, it appears that predator species are able to co- occur in many different combinations, creating a range of phylogenetic diversity found within bromeliads. This suggests that the range of phylogenetic diversity we tested in our experiment was not unrealistic. It also indicates that whatever the effects of interactions between predator species, they are not sufficient to limit species distributions.

### Phylogenetic distance and similarity in diet

We observed a negative relationship between phylogenetic distance and overlap in diet as measured by laboratory feeding trials. These extreme differences were likely due to phylogenetically-distant predator species possessing very different feeding habits. For example, damselflies are visual predators () which consume prey whole using specialized mouthparts; they are gape-limited and cannot eat prey that is too large. Leeches, in contrast lack eyes but are able to pierce prey and consume it without swallowing. Damselflies showed a much stronger preference for culicid larvae than did leeches, while leeches were slightly more able to kill and eat scirtids. Such traits can be more important than phylogeny *per se* to a predator's diet: (Moody 1993) found that unrelated decapod species which were morphologically similar were also functionally similar. Similarly, (Rezende et al. 2009) found that both body size and phylogeny determined the food web "compartment" (shared predator-prey interactions) of a predator in a marine foodweb. In addition, our experimental results are consistent with high similarity among predator diets: all predator species had comparable effects on all response variables, including prey species survival.

### Phylogenetic diversity and non-additive effects

We found that the presence of predators reduced prey survival, but that this reduction was less for more phylogenetically-diverse combinations of predators. This is contrary to our hypothesis that more distant predators would show an increase in prey capture via complementarity. *L. andromache* did not produce a negative effect in combination with *L. elongatum*, while more phylogenetically distinct predators did. It may be that these odonates have behavioural traits that reduce the probability of their interaction, for example each nymph occupying a single leaf-well. If this is the case, each damselfly may not experience many cues indicating the presence of other predators, resulting in no non- additive interaction.

We also found that the increase in prey survivorship was caused by a negative non-additive effect of predator species. The principle cause of the negative non-additive effect may have been a reduction in predation by odonates. Odonates can be sensitive to chemical cues of potential predators, which causes a decrease in feeding rate (Barry and Roberts 2014). However other research in bromeliads has demonstrated that it is physical contact with other organisms that reduces damselfly predation rate (). This may be the mechanism for reduced predation by *Leptagrion* when combined with leeches, as leeches are very active and could make frequent physical contact. Tabanids, however, are rarely observed outside of a deep leaf axil -- in this case, it may be chemical cues which are responsible. There may also be a phylogenetic signal to the chemical cues which the damselflies perceive: i.e. close relatives (other *Leptagrion* sp. in this case) might induce less of an effect than other predators. One limitation of our approach is that all phylogenetic diversity treatments contained one species in common, *Leptagrion elongatum*. It is possible that this species is more sensitive to the presence of other predators, and therefore shows a larger trait-mediated indirect effect, than would other species in this community. However, this is the most common species in this community and our results indicate that its top-down effects are likely to be frequently reduced by the presence of other predators.

In most natural communities, diverse predator species co-occur and often simultaneously affect prey species. There is no clear way to predict how these diverse combinations will influence the structure and functioning of lower trophic levels. An approach based on phylogenetic diversity offers an organizing framework around which to compare diverse datasets on the distribution, trophic interactions and combined effect of multiple predator species, potentially leading to a means of predicting the top-down effect of diverse predator assemblages.

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