Trophic interactions modify the temperature dependence of community biomass and ecosystem function

Running head: ecological effects of temperature

Jessica Garzke[*1,2], Stephanie J. Connor[3], Ulrich Sommer[1] and Mary I. O'Connor[4]

1 Geomar Helmholtz Centre for Ocean Research Kiel, Dept. Experimental Ecology – Food Webs, Duesternbrooker Weg 20, DE-24105 Kiel, Germany

2 Institute of the Oceans and Fisheries, University of British Columbia, Vancouver, Canada, V6T 1Z4

3 Canadian Rivers Institute, Dept. of Biology, University of New Brunswick, Fredericton, Canada, E3B 5A3

4 Department of Zoology and Biodiversity Research Centre, University of British Columbia, Vancouver, Canada, V6T 1Z4

Statement of authorship: JG & MIO designed the study, MIO & US provided materials, JG & SJC performed research and collected data, JG performed zooplankton analysis, SJC performed phytoplankton analysis, JG & MIO performed modeling work, analyzed data output, and wrote the first draft, and all authors contributed substantially to reviews

***Corresponding author:** Jessica Garzke, j.garzke@oceans.ubc.ca, +1 778-772-6977, ORCID: 0000-0002-5881-1813

Keywords: community structure, metabolic theory of ecology, zooplankton, phytoplankton, temperature size rule, mesocosm experiment, ecosystem respiration, net primary production

Research paper

Abstract

Aquatic ecosystems worldwide continue to experience unprecedented warming and ecological change. Warming increases metabolic rates of animals, plants and microbes, accelerating their use of energy and materials, their population growth and interaction rates. At a much larger biological scale, warming accelerates ecosystem level processes, elevating fluxes of carbon and oxygen between biota and the atmosphere. Though these general effects of temperature at finer and broader biological scales are widely observed, they can lead to contradictory predictions for how warming affects the structure and function of ecological communities at the intermediate scale of biological organization. We experimentally tested the hypothesis that the presence of predators and their associated species interactions modify the temperature-dependence of net ecosystem oxygen production and respiration. We tracked a series of independent freshwater ecosystems (370-L) over 9 weeks, and we found that at higher temperatures, cascading effects of predators on zooplankton prey and algae were stronger than at lower temperatures. When grazing was weak or absent, standing phytoplankton biomass declined by 85-95% (< 1-fold) over the temperature gradient (19-30 °C), and by 3-fold when grazers were present and lacked predators. Surprisingly, these temperature-dependent species interactions and consequent community biomass shifts occurred without signs of species loss or community collapse, and only modestly affected the temperature dependence of net ecosystem oxygen fluxes. The exponential increases in net ecosystem oxygen production and consumption were relatively insensitive to differences in trophic interactions among ecosystems. Furthermore, monotonic declines in phytoplankton standing stock suggested no threshold effects of warming across systems. We conclude that local changes in community structure, including temperature-dependent trophic cascades, may be compatible with prevailing and predictable effects of temperature on ecosystem functions related to fundamental effects of temperature on metabolism.

Introduction

Temperature affects metabolic rates of all organisms, thereby affecting ecological patterns and processes across scales of organization – from individuals to ecosystems. Increasing temperature accelerates major metabolic processes that drive net ecosystem production and ecosystem respiration in aquatic and terrestrial ecosystems [1-4]. Highly conserved metabolic processes - photosynthesis and aerobic respiration [5] - power somatic growth, maintenance and activity in aerobic organisms. As a result, the effects of temperature on cellular photosynthesis and respiration have accurately described the exponential increases in ecosystem-scale ecosystem productivity (NEP) and respiration (ER) in aquatic systems across macroecological thermal gradients, after accounting for body size, nutrient content and light availability [4,6,7]. The ecological importance of temperature-dependent *per capita* metabolic rates has supported to the use of metabolic models, and the development of an important theme in the metabolic theory of ecology (MTE) of general temperature dependence, to understand and predict ecological change from local to global scales [3,4,8]. Models that associate change in ecosystem scale metabolism (e.g., oxygen or carbon flux) with individual-level oxygen production and respiration, but bypass the complexity of population and community dynamics at intermediate biological scales, provide much needed predictability for how climate change affects ecosystem functions when ecosystems are compared across broad spatial or temporal thermal gradients [2,4,9].

Reconciling the high explanatory power of general temperature-dependent metabolic scaling models at macroecological scales with the well-documented contingencies of how temperature affects community level outcomes of population dynamics and species interactions at intermediate scales has been challenging [10-17]. Whether at macro-ecological or community (e.g., single-site) scales, ecosystem-level functions (ER, NEP) or biomass is simply the sum of per capita function (respiration, net photosynthesis) and biomass. Metabolic theory models applied at macro-ecological scales assume that the relationship

between temperature and community-level distributions of body sizes and traits is constant in time, or that communities are at stable state so that descriptions of community structure apply to future states of the community under the same abiotic conditions [10-12]. Yet, at local scales, species interactions can influence biomass of primary producers, and the strength and outcomes of species interactions reflect dynamical processes that are often sensitive to temperature [13-15]. For example, the presence of fish in experimental aquatic ponds reversed a negative effect of temperature on algal biomass to a positive effect, mediated by trophic interactions between fish, zooplankton and phytoplankton [16], under otherwise constant consistent abiotic conditions across ponds. Understanding the temperature dependence of species interactions and their consequences for how biomass, size distributions and traits vary under even constant abiotic conditions raises a challenge for the application of general temperature dependence models that assume individual metabolism to change at community scales without explicitly measuring [17,18]. This paradox between macroecological patterns – which can be consistent with direct scaling of per capita thermal responses - and results of smaller scale, short term experiments that allow population dynamics to play out over intermediate time scales, leads to the suggestion that general metabolic scaling models that do not consider the complexities associated with species interactions do not apply at the local scales [19]. Reconciling these apparently divergent patterns is critical to improving understanding and projections of how shifting global thermal regimes affect ecological patterns and processes across scales and achieving a more unified understanding of ecology across scales.

One way to reconcile the apparent context dependence of empirical results under controlled conditions with the generality of temperature dependence of ecosystem function at broader scales is to consider how the direct and indirect effects of temperature on population dynamics interact. Direct effects of temperature on *per capita* metabolic rates cause organismal photosynthesis and respiration rates to increase exponentially as temperatures

increase when resources are not limiting in algae and animals, up to an optimal temperature. This relationship between temperature and fundamental metabolic rates (photosynthesis and respiration) is referred to as general metabolic scaling [1]. For any single phenotype, performance above some optimal temperature declines due to stress responses and metabolic scaling no longer explains the effects of temperature on perfromance. However, in multi-species communities the signal of metabolic scaling is likely to dominate over a broad range of temperatures if species with distinct thermal phenotypes can compensate for each other along the thermal gradient [10,20]. Warming is also associated with other biological changes that affect species interactions, such as reductions in body size (the temperature size rule, [21-23]), fecundity, and attack rates (Fig 1) [24-27], and these changes can feed back to influence community-level biomass and productivity [28-30].

Figure 1. A) Temperature and predation directly and indirectly affect population density and metabolic rates in aquatic communities. In our experimental communities, predation directly (solid lines) affects the abundance, size and species composition of prey, and predation by notonectids on grazers leads to an indirect effect (dashed line) called a trophic cascade on algal abundance. Temperature directly affects per capita metabolic rates (solid lines), and indirectly affects algal abundance (dashed lines) by increasing grazing rates. Other indirect effects of temperature are possible, but not shown. B) Experimental communities varied in their trophic structure. Ten communities included algae only (A), 10 comprised algae + grazers (AG), and 10 included algae + grazers + predators (AGP). We sampled net ecosystem oxygen production (NEP), ecosystem respiration (ER) and total phytoplankton biomass (M_B) weekly for 8 weeks.

The temperature-dependence of consumer-resource interactions – mediated by

129 dynamics of two or more populations - has received substantial attention in this context,
130 because these *trophic* interactions can influence many aspects of community structure and
131 ecosystem function, including biomass, abundance, species composition and stability
132 [10,12,32,34]. Trophic species interactions appear to strengthen with warming [16,29,31].
133 Series of trophic interactions, called *trophic cascades* (Fig 1), link predator populations to the
134 abundance, biomass and ecosystem functions of primary producers [32,33] and the strength of
135 trophic cascades depends on body size and primary production [34]. Considering the
136 multitude of indirect effects of temperature on population dynamics and the prevalence of
137 consumer-resource interactions and trophic cascades in aquatic systems [35] begs the
138 question, how is it that these locally dominant population-level responses to temperature
139 [28,36] do not appear to cause major variation or context dependence in macroecological
140 relationships between subcellular metabolic processes (photosynthesis, respiration) and
141 ecosystem processes (NEP, ER)?
142 Here, we aimed to resolve the paradox between apparent direct effects of temperature
143 on ecosystem functions (NEP, ER) that emerge when comparing communities across larger
144 gradients and the potentially more complex effects of temperature at the population and
145 community scales. We experimentally tested the hypothesis that temperature-dependent
146 trophic interactions in a trophic cascade alter the effect of temperature on community
147 properties such as biomass, abundance and body size, but have little or no effect on the
148 temperature dependence of ecosystem functions (NEP, ER) over a temperature gradient. In
149 freshwater plankton communities, we compared the effects of temperature on community
150 properties typically measured in warming experiments (e.g., biomass, density, body size) with
151 the effects of temperature typically compared in macroecological studies (e.g., NEP, ER). We
152 controlled variation in biotic and abiotic conditions other than temperature and trophic
153 structure (presence of grazers and predators) (Fig 1). We quantified ecosystem function
154 (NEP, ER) and community structure (biomass, abundance) in ecosystems with algae only (A),

algae and grazers (AG), or algae, grazers and predators (AGP) across an experimental temperature gradient of 10 °C. We found that exponential effects of temperature on algal biomass were greater than effects of temperature on NEP and ER, suggesting that even large changes in community structure do not necessarily lead to large changes in how temperature affects NEP and ER.

Hypotheses

We drew on the Metabolic Theory of Ecology (MTE) frame our hypotheses and predictions for how temperature affects NEP and ER via *per capita* metabolic temperature dependence and indirect effects of temperature at the community scale. We first briefly outline the framework, and then express our specific hypotheses. MTE relates whole-organism metabolic rates (b_i, gO_2/hr) and related biological functions for organism *i* to body size (m_i, g) and body temperature (T, in Kelvin) [1,37,38]:

$$b_i = b_0 e^{-E_a/kT} m_i^a \qquad \text{Eqn 1}$$

in which activation energy (E_a, in eV) captures the exponential effect of temperature on per capita metabolic rate, k is the Boltzmann constant (eV/K), b_0 is a normalization constant independent of body size and temperature that includes the effects of temperature-independent traits on metabolic rate (gO_2/g^{α}/hr). The allometric scaling factor α relates metabolic rate to body size [see Methods: Models and Hypotheses for justification of Eqn 1 over alternative models].

The effects of temperature on ecosystem metabolic rates (B_R), such as NEP or ER (gO_2/hr) reflect the sum of all *per capita* photosynthesis rates by autotrophs and respiration rates by autotrophs and heterotrophs as well as shifts in abundance, body size and acclimation. These models implicitly assume ample and constant supply of resources. Note that NEP and ER can be quantified in this way as positive numbers, and we do this – using their absolute values – in our analyses. Following Barneche et al (2014), we capture direct

and indirect effects of temperature on ecosystem scale metabolic rates in the following equation (see Barneche et al 2014 for derivation):

$$B_R = b_0(T_C)\, e^{-E_R\left(1/kT - 1/kT_C\right)}\, M_B\langle m_B^{\alpha-1}\rangle. \qquad \text{Eqn 2}$$

The term $e^{-E_R\left(1/kT - 1/kT_C\right)}$ captures the temperature dependence E_R (eV) of ecosystem-level metabolic rate B_R. Equation 2 represents a 'first order metabolic scaling' prediction that ecosystem scale mass-normalized metabolic rates (e.g., NEP) vary proportionally with the temperature dependence of the underlying metabolic processes (e.g., photosynthesis). Observed temperatures T are related to an arbitrarily chosen reference temperature T_c. This centering causes the normalization constant $b_0(T_C)$ to be for metabolic performance at temperature T_c.

When considering the indirect effects of temperature on ecosystem oxygen production and respiration, we can consider how each term in Eqn 2 may vary with temperature. To account for changes in total biomass, body size or relative abundance of phenotypes (traits) associated with temperature, we use the term $M_B\langle m_i^{\alpha-1}\rangle_B$. The total biomass, M_B (g) in ecosystem volume V, which equals the sum of mass m_i for all individuals i to J ($M_B = \frac{1}{V}\sum_{i=1}^{J} m_i$). The term $\langle m_i^{\alpha-1}\rangle$ is the average of all individual metabolic biomasses, $\langle m_i^{\alpha-1}\rangle = \left(\sum_{i=1}^{J} m_i^{\alpha}\right)/\left(\sum_{i=1}^{J} m_i\right)$, corrected for the greater contribution to total mass-specific metabolic biomass by small individuals resulting from the allometric scaling (α) of oxygen production and consumption with body size [2,10]. This 'mass correction' is necessary, because if community biomass is comprised of one large individual, that biomass will [produce and] consume less oxygen per gram biomass in a given time period than if the same total biomass were comprised of many small individuals; in other words, $M_B\langle m_B^{\alpha-1}\rangle$ increases as m_i. If thermal traits acclimate or species composition shifts with temperature, this term would capture that change. Therefore Eqn 2 can capture direct effects of temperature on community metabolism via changes in per capita metabolic rate (E_R) and via changes in biomass, size

distribution and phenotypes.

Hypothesis 1: Trophic interactions modify the effect of temperature dependent metabolic rates on total algal biomass. Via strong trophic interactions, predators can change the standing biomass of primary producers in communities. Total algal biomass (M_B) can be expressed in terms of temperature, traits and size distributions:

$$M_B = \frac{B_R\, e^{E_R\left(1/kT^{-1}/kT_C\right)}}{b_0(T_C)\langle m_B^{\alpha-1}\rangle}. \qquad \text{Eqn 3}$$

If we assume that $B_R * (b_0(T_C) * \langle m_B^{\alpha-1}\rangle)^{-1}$ is independent of temperature, we predict that algal biomass M_B declines with temperature by E_R, in this case $E_R = -E_{NEP}$. This prediction has been supported empirically in a single species algae system [39], and in that system the predicted decline in total biomass was robust to changes in cell size. However, it is unlikely that grazers and temperature would not alter the abundance and size of algae, altering $\langle m_B^{\alpha-1}\rangle$ among trophic treatments [40], and also the traits of algae, and thereby modifying $b_0(T_C)$ among trophic treatments [41]. A fuller integration of how temperature and trophic treatment affect these terms for multispecies assemblages would require theoretical development that is beyond the scope of this paper, but we use the equation here to highlight why we expect trophic structure and temperature to affect algal biomass. To test this hypothesis, we linearized Eqn 3 for analysis by log transforming (Methods: Eqn 7) and then we compared ln(M_B) trends with temperature across ecosystems with and without a trophic cascade (AGP vs AG ecosystems).

Hypothesis 2: Increasing temperature strengthens the trophic cascade. We estimated the strength of the trophic cascade as the log ratio of primary producer biomass in the presence of predators (AGP) *vs* in predator-free environments (AG) [42]. We predicted that predators would reduce the abundance of zooplankton through predation, and shift zooplankton

231 composition to smaller sizes and less-edible species, typical of classic freshwater trophic
232 cascades [43], and that these trophic interactions would strengthen with higher temperatures
233 due to the effect of temperature on *per capita* grazing rates. We can relate algal biomass
234 among treatments using Eqn 3 for primary producer biomass in the presence of predators
235 (AGP) and grazers only (AG), simplifying and taking the natural log to yield (see Methods,
236 eqns 7-10, for details):

237 $$ln\left(\frac{M_{B.AGP}}{M_{B.AG}}\right) = \frac{\ln(b_0(T_C)_{AG}) + ln\left(\langle m_B^{\alpha-1}\rangle_{AG}\right) - \frac{E_{b.ag} - E_{m.ag}}{kT}}{\ln(b_0(T_C)_{AGP}) + ln\left(\langle m_B^{\alpha-1}\rangle_{AGP}\right) - \frac{E_{b.agp} - E_{m.agp}}{kT}}$$ Eqn 4

238 Numerous experiments have demonstrated that the strength of the trophic cascade (log $\frac{M_{B.AGP}}{M_{B.AG}}$,
239 increases with increases in temperature of a few degrees [29,44,45], and theoretical work
240 suggests that strengthening of this interaction under warming is expected for a greater range
241 of consumer-resource parameter values than would predict declines in the trophic cascade
242 [15]. We therefore predict the trophic cascade strength will increase with temperature in our
243 experiment. But, as shown in Eqn 4, because trophic interactions could affect the realized
244 temperature dependence via several possible mechanisms – shifts in body size, traits, etc, - it
245 is not possible to predict *a priori* the temperature dependence of the trophic cascade, in terms
246 of the differences and ratios of the temperature dependence terms in Eqn 4 (see Methods). A
247 'first order' metabolic scaling prediction would be no change in trophic cascade strength,
248 because the model would assume that the temperature dependences of mass and the
249 normalization constant ($E_{b.ag}$, $E_{b.agp}$, $E_{m.ag}$, and $E_{m.agp}$) all equal 0. We expected the indirect
250 effects of predators on algae to be mediated by changes in zooplankton density and/or body
251 size. Zooplankton attributes are not explicitly modeled in Eqn 4, but could contribute to
252 temperature dependence of algal cell size and trait distributions. Reduced zooplankton size or
253 density in the presence of predators could lead to different indirect effects of temperature on
254 algal cell size and traits in the presence vs absence of predators. We tested this expectation by
255 testing the prediction that temperature dependence of zooplankton size and density are

different from zero (Methods: Statistical Analysis).

Hypothesis 3: Temperature dependence of net ecosystem production and respiration depends on the strength of the trophic cascade. We test this by using Equation 2 to model ecosystem-scale NEP and ER, but we allow $b_o(T_C)$ to vary not only with temperature but with trophic structure (Z_j). We expect that trophic structure will influence the number and size of individuals, and thereby affect $M_B\langle m_B^{\alpha-1}\rangle$. Alternatively, trophic structure may not modify the relationship between temperature and B_R, if $b_o(T_C)$ and $\langle m_B^{\alpha-1}\rangle$ are independent of temperature. We can test these alternate predictions by comparing models with and without $b_o(T_C)$ and $\langle m_B^{\alpha-1}\rangle$ terms that depend on ecosystem temperature and trophic structure.

For each hypothesis, we used linear mixed effects models (LMMs) to test 'first order' metabolic scaling models for the appropriate model (equation 2, 3 or 4) that included $b_o(T_C)$ as independent of the ecosystem's temperature or trophic structure [Methods: Statistical Analyses]. We tested alternate models that allowed $b_o(T_C)$ to vary with ecosystem temperature and/or trophic structure. If the simpler, first order models are best supported, we would infer that indirect effects of temperature do not overwhelm the signals of direct metabolic scaling effects on ecosystem functions, consistent with inferences drawn in macroecological studies. To estimate intercepts and temperature dependence terms (e.g., E_R), we summed coefficient values and estimated uncertainties in these aggregated parameters from best models (Methods: Statistical Analysis). For each hypothesis, we tested two measures of ecosystem temperature: mean temperature over the 9-week experiment, which captures differences among systems, or weekly mean temperature, which captures differences within ecosystems over time. Our data do not permit testing specific predictions about size distributions or trait shifts, but support for models with variation in $b_o(T_C)$ and $\langle m_B^{\alpha-1}\rangle$ among treatments would suggest these mechanisms as likely explanations.

Results

Hypothesis 1: As temperature increased across ecosystems, phytoplankton biomass, estimated as the concentration of chlorophyll *a* in the water column, declined (Fig 2A). Trophic interactions modified the effect of temperature on chlorophyll *a* concentration (Fig 2A, Table 1). This inference is supported by the inclusion of a main effect for trophic structure (Z_j) in the best model (Table 1) and an estimate for the temperature dependence of chlorophyll *a* concentration with confidence intervals that exclude 0 (Fig 3). Phytoplankton biomass declined much more strongly with temperature in algae-grazer (predator-free) communities, with a decline of over three orders of magnitude in phytoplankton biomass standing stock over the 10 °C temperature gradient (Fig 2A). In the algae-grazer-predator treatments, phytoplankton biomass declined with a slope indistinguishable from that in the algae only treatments (Fig 3). We did not observe shifts in taxonomic composition with temperature (Fig S3).

Figure 2: A) Estimated phytoplankton biomass (chlorophyll *a* concentration) declined with increasing temperature and varied with trophic structure (A, AG, AGP). Lines are estimated effects of temperature on phytoplankton biomass based on linear mixed effects models (Eqn 11) for equation 3 with temperature dependent in model terms for the intercept and slope (Table 1). From the best model, the intercept and slope of each line was estimated by pooling terms for the intercept and temperature dependence in eqn 11 (see Methods, Eqn 13). All observations for phytoplankton biomass are shown in Fig 6. **B**) Strength of the trophic cascade at a given temperature was estimated by taking the log ratio of algal biomass (estimated as chlorophyll *a* concentration) in the presence of predators and grazers (AGP) vs the algal abundance in the presence of grazers only (AG) (Eqn 4, Table 2). Lines represent

fixed effects of temperature from the full model (Table 2), centered on the grand mean of all recorded ecosystem temperatures (Eqn 12).

Table 1. Model selection results for linear mixed effects models of phytoplankton biomass. The full statistical model (Methods: Eqn 11) related ln(chlorophyll *a*) to ecosystem trophic structure (Z_j) and average ecosystem temperature over the entire experimental period (T_M), while accounting for effects of temperature variation over time (weekly average temperature (T_{wj})) and with ecosystem identity as a random effect. We compared models using likelihood ratios (LogLik), AICc, Akaike weights (w) and delta AICc weights. The model was fit to 240 observations in 30 groups. The full model (modelPBF) includes all terms, and models representing alternate hypotheses excluded terms indicated by 'NA'. Values indicate model-estimated coefficients. Coefficients were pooled (Methods: Statistical Analysis) to estimate slopes and intercepts for Fig 2 and 3.

	Int	Z_j	T_{wj}	T_M	T_{wj}*Z_j	T_M*Z_j	T_M*T_{wj}	df	logLik	AICc	d	w
modPBF	2.05	+	-0.52	1.30	1.34	+	+	12	-155.37	336.11	0.00	9.982260e-01
modPB8	2.05	+	-0.66	1.30	NA	+	+	11	-162.86	348.87	12.76	1.690617e-03
modPB7	2.05	+	-0.96	1.30	NA	NA	+	9	-168.05	354.89	18.78	8.342130e-05
modPB4	1.50	NA	-0.96	1.70	0.96	NA	NA	6	-207.94	428.24	92.13	9.830504e-21
modPB6	1.91	+	-0.66	NA	NA	+	NA	8	-206.58	429.79	93.68	4.538012e-21
modPB3	1.50	NA	-0.96	1.71	NA	NA	NA	5	-211.73	433.72	97.62	6.341539e-22
modPB5	1.91	+	-0.96	NA	NA	NA	NA	6	-211.45	435.26	99.16	2.937427e-22
modPB2	1.50	NA	-0.96	NA	NA	NA	NA	4	-218.40	444.97	108.87	2.287011e-24
modPB1	1.90	+	NA	NA	NA	NA	NA	5	-257.21	524.68	188.57	1.124143e-41
modPB0	1.49	NA	NA	NA	NA	NA	NA	3	-264.15	534.41	198.30	8.684503e-44

Figure 3. Comparison of estimated temperature dependences of phytoplankton biomass (M_B), net ecosystem oxygen production (NEP), ecosystem respiration (ER) for communities with algae only (A), algae and grazers (AG) and algae, grazers and predators (AGP). Composite estimates of temperature dependences as shown in Figures 2A and 3 (following Methods, Eqn 13). No temperature dependence is indicated by the dashed line, and the gray dotted lines indicate $\pm$ 0.65 and 0.32 eV, expected temperature dependences of photosynthesis and

respiration, and -0.65 and -0.32 as expectations for the temperature dependence of phytoplankton total biomass.

Hypothesis 2: Consistent with our second hypothesis, and the patterns observed for phytoplankton biomass, there was a strong trophic cascade in the warm ecosystems by the end of the experiment (Fig 2B). The trophic cascade became apparent after the first weeks of the experiment, and strengthened over time and with temperature (Fig 2B) (Table 2). The best model included a term for mean ecosystem temperature (T_M), as well as week (T_w), and a week x temperature interaction. By week 9, the trophic cascade increased exponentially with temperature (Fig 2B) to an estimated E_{TC} = 0.77 (estimated from model fixed effects shown in Table 2 plus random effect, not shown).

Table 2. Model selection results for trophic cascade analysis. We used linear mixed effects models with terms for average temperature for ecosystem *j* in week *w* (T_{wj}), week 2-9 (Wk) and their interaction. We treated the power level (e.g., 100W, 200W, etc), our temperature treatment, as a random effect to account for repeated measures on ecosystems over time. We compared models using likelihood ratios (LogLik), AICc, Akaike weights (w) and delta AICc weights. The model was fit to 79 observations in 10 groups. The full model (TCFull) includes all terms, and models representing alternate hypotheses excluded terms indicated by 'NA'. Coefficients were pooled (Methods) to estimate slopes and intercepts for Fig 2.

	Int	T_{wj}	Wk	T_{wj}*Wk	df	logLik	AICc	d	w
TCFull	0.19	-0.01	0.12	0.11	6	-60.78	134.73	0.00	0.769299027
TCmodc	0.02	0.74	0.14	NA	5	-63.26	137.33	2.60	0.209484257
TCmode	0.56	NA	0.04	NA	4	-67.36	143.27	8.54	0.010776915
TCmodf	0.79	NA	NA	NA	3	-68.80	143.92	9.19	0.007786805
TCmodd	0.79	-0.05	NA	NA	4	-68.76	146.07	11.34	0.002652997

We find additional evidence of temperature-dependent trophic interactions in the responses of the zooplankton grazer assemblages to warming and predation. Total zooplankton density declined with increasing temperature (Table 3; E_{ZP} = 1.28 95% CI: 0.19 – 2.39, z = 2.31, P = 0.021 based on regression with negative binomial distribution and likelihood ratio tests; Fig 4). Predators reduced density of *Daphnia*, the dominant grazer (linear regression with Poisson distributed errors: estimate: = -1.14 95% CI: -1.91, -0.36, z = -2.87, $P < 0.01$) and density declined with increasing temperature (Table 4) although the temperature term retained in the best model was not significant (E_D = 0.88 95% CI: -0.59, 2.35, z = 1.17, P = 0.24). Copepod density declined with temperature (E_C = 2.21, z = 3.12, P = 0.002), and not in response to predation (best model did not include a predation term, Table 6). We measured zooplankton standard length for 641 individuals of all ages. Mean length was 0.72 cm, and sizes ranged from 0.34 – 1.94 cm. We did not observe a decline in body size with temperature (best model did not include a temperature term, Table S2), as would be expected by a hypothesis based on the temperature size rule. Predation reduced total zooplankton body size, driven by size shifts in Daphnia (Table S3).

Table 3: Zooplankton density. Results of model selection for zooplankton abundance in ecosystems with grazers (AG) and with grazers and predators (AGP). We used negative binomial regressions with ecosystem identity as a random effect [Methods: Statistical Analysis]. Models included terms for weekly average temperature (T_{wj}), ecosystem trophic treatment (Z_j) and their interaction, and a random effect for ecosystem identity. We modeled 120 observations in 20 groups (ecosystems). We compared models using likelihood ratios (LogLik), AICc, Akaike weights (w) and delta AICc weights. NA indicates that the term was not included in the model.

	Int	T_{wj}	Z_j	df	logLik	AICc	d	w
nbinommod1b	-47.41	1.28	NA	5	-367.00	744.53	0.00	0.49557915
nbinommod1	-48.76	1.32	+	6	-366.37	745.47	0.95	0.30895745
nbinommod1c	2.95	NA	NA	4	-369.46	747.26	2.74	0.12621312
nbinommod1a	3.07	NA	+	5	-368.97	748.46	3.94	0.06925027

Table 4: Daphnia density: Results of model selection for *Daphnia* abundance in ecosystems with grazers and with grazers and predators. We used Poisson regressions with ecosystem identity as a random effect [Methods: Statistical Analysis]. Models included terms for weekly average temperature (T_{wj}), ecosystem trophic treatment (Z_j) and their interaction, and a random effect for ecosystem identity. We compared models using likelihood ratios (LogLik), AICc, Akaike weights (w) and delta AICc weights. We modeled 120 observations in 20 groups (ecosystems). NA indicates that the term was not included in the model.

	Int	T_{wj}	Z_j	T_{wj}*Z_j	df	logLik	AICc	d	w
poismod.Db	-34.88	0.88	+	NA	4	-115.75	239.85	0.00	0.63118106
poismod.Da	-38.64	0.97	+	+	5	-115.73	241.98	2.13	0.21709779
poismod.Dd	-0.97	NA	NA	NA	2	-119.71	243.52	3.67	0.10060719
poismod.Dc	-31.32	0.77	NA	NA	3	-119.33	244.87	5.03	0.05111395

Table 5: Copepod density: Results of model selection for copepod *spp* abundance in ecosystems with grazers and with grazers and predators. We used Poisson regressions with ecosystem identity as a random effect [Methods: Statistical Analysis]. Models included terms for weekly average temperature (T_{wj}), ecosystem trophic treatment (Z_j) and their interaction, and a random effect for ecosystem identity. We compared models using likelihood ratios (LogLik), AICc, Akaike weights (w) and delta AICc weights. We modeled 120 observations in 20 groups (ecosystems). NA indicates that the term was not included in the model.

	Int	T_{wj}	Z_j	T_{wj}*Z_j	df	logLik	AICc	d	w
poismod.Cc	-87.63	2.21	NA	NA	4	-135.11	278.56	0.00	0.67753474
poismod.Cb	-87.50	2.21	+	NA	5	-135.10	280.72	2.16	0.23026158
poismod.Ca	-96.20	2.43	+	+	6	-135.04	282.83	4.27	0.07997327
poismod.Cd	-0.34	NA	NA	NA	3	-140.19	286.59	8.03	0.01223041

Figure 4: A) Total zooplankton density (ind / 10L), comprising *Daphnia* and copepod taxa, declined with increasing temperature but not with predator presence. B) *Daphnia* density (ind/L) declined with temperature and with predators (Table 4), and C) copepod spp density (ind/L) declined with temperature but not predators (Table 5). Lines are regression fits with negative bionomial (total zooplankton) or Poisson (Daphnia, copepods) zero-inflated error structures with ecosystem as a random effect for ecosystems with predators (gray dashed line) and without predators (black solid line). Each datapoint is an observed total zooplankton density for crustacean taxa (*Daphnia* and copepods) in each ecosystem on a sampling date.

Hypothesis 3: Across ecosystems higher temperatures increased net ecosystem oxygen production (NEP) and respiration (ER) (Tables 6, 7; Fig 5). The lmm for NEP (Table 6) suggests that ecosystem temperature and trophic structure interact to influence ecosystem oxygen fluxes, yet their estimated temperature dependences did not appear to differ when confidence intervals were compared (Fig 3). The estimated across-system temperature dependence of NEP was the strongest in algae-only communities (Fig 5), and confidence intervals for the temperature dependence term include 0 for the systems with predators (Fig 3). Ecosystem respiration (ER) increased with temperature across ecosystems (Fig 5), and this effect did depend on trophic structure (Table 7). The estimated temperature dependence on NEP and ER was strongest in the algae-only systems, and weakest in systems with predators (Fig 3).

Table 6: Results of model comparisons for effects of temperature and time on net ecosystem productivity (NEP) based on AIC weight (*w*) and δAIC_C values. Nested versions of the full model (Eqn 11, Methods). Response variables are modelled as functions of temperature T_{wj} for each tank *j* on week *w* relative to the mean temperature T_M for tank *j* over all weeks (T in Kelvin), and trophic structure (Zj). Models included a random effect for the experimental unit – tanks with and without predators receiving the same power inputs. See Methods for additional details on modeling. The model was fit to 219 observations in 30 groups. The full model (modNEPF) includes all terms, and models representing alternate hypotheses excluded terms indicated by 'NA'. Values indicate model-estimated coefficients. Coefficients were pooled (Methods: Statistical Analysis) to estimate slopes and intercepts for Fig 3 and 5.

	Int	Z_j	T_{wj}	T_M	T_{wj}*Z_j	T_M*Z_j	T_{wj}*T_M	df	logLik	AICc	d	w
modNEP8	-6.42	+	0.29	-1.40	NA	+	+	11	-266.47	556.21	0.00	3.878747e-01
modNEPF	-6.42	+	0.37	-1.42	0.84	+	+	12	-265.54	556.60	0.39	3.192963e-01
modNEP7	-6.41	+	0.03	-1.39	NA	NA	+	9	-269.68	558.22	2.01	1.421535e-01
modNEP3	-6.15	NA	0.02	-0.96	NA	NA	NA	5	-274.36	559.01	2.80	9.557409e-02
modNEP4	-6.15	NA	0.02	-0.96	0.61	NA	NA	6	-273.86	560.12	3.91	5.487563e-02
modNEP0	-6.15	NA	NA	NA	NA	NA	NA	3	-283.15	572.41	16.20	1.179389e-04
modNEP2	-6.15	NA	0.03	NA	NA	NA	NA	4	-283.13	574.44	18.23	4.265400e-05
modNEP1	-6.26	+	NA	NA	NA	NA	NA	5	-282.25	574.78	18.57	3.596974e-05
modNEP6	-6.26	+	0.27	NA	NA	+	NA	8	-279.83	576.34	20.13	1.647426e-05
modNEP5	-6.26	+	0.03	NA	NA	NA	NA	6	-282.23	576.85	20.64	1.278556e-05

Table 7: Results of model comparison for effects of temperature and time on ecosystem respiration (ER) based on AIC weight (w) and δAIC_C values. Nested versions of the full model (Eqn 11, Methods). Response variables are modelled as functions of temperature T_{wj} for each tank *j* on week *w* relative to the mean temperature T_M for tank *j* over all weeks (T in Kelvin), and trophic structure (Z_j). Models included a random effect for the experimental unit – tanks with and without predators receiving the same power inputs. The model was fit to 240 observations in 30 groups. See Methods for additional details on modeling.

	Int	Z_J	T_{wJ}	T_M	T_{wJ}*T_M	T_{wJ}*Z_J	T_M*Z_J	df	logLik	AICc	d	w
modER7	-6.09	+	0.11	-1.32	NA	NA	+	9	-185.88	390.54	0.00	6.013204e-01
modER8	-6.09	+	0.02	-1.32	NA	+	+	11	-184.58	392.31	1.77	2.479721e-01
modERF	-6.09	+	0.06	-1.32	0.42	+	+	12	-183.97	393.31	2.77	1.506867e-01
modER3	-5.79	NA	0.11	-0.67	NA	NA	NA	5	-201.37	413.00	22.46	7.978045e-06
modER4	-5.79	NA	0.11	-0.68	0.50	NA	NA	6	-200.45	413.27	22.73	6.977684e-06
modER1	-5.94	+	NA	NA	NA	NA	NA	5	-202.46	415.18	24.64	2.685333e-06
modER5	-5.94	+	0.11	NA	NA	NA	NA	6	-201.75	415.85	25.31	1.916292e-06
modER6	-5.94	+	0.02	NA	NA	+	NA	8	-200.49	417.60	27.06	8.013775e-07
modER0	-5.79	NA	NA	NA	NA	NA	NA	3	-207.04	420.17	29.63	2.211271e-07
modER2	-5.79	NA	0.11	NA	NA	NA	NA	4	-206.32	420.82	30.28	1.602501e-07

Figure 5: The effect of mean ecosystem temperature on A) phytoplankton biomass, B) net ecosystem productivity (NEP), and C) net ecosystem respiration (ER) for three community types that varied in their trophic interactions: i) algae-only (A), ii) algae + grazers (AG), and iii) algae + grazers + notonectid predators (AGP). Black lines indicate the among-ecosystem effects of temperature, modelled by equation 5 using hierarchical regressions fit to among-ecosystem variation in temperature, after taking into account within-group variation temperature effects (light lines) (Table 2), and may be compared with predicted effects of temperature and species interactions depicted in Figure 1. Activation energies and confidence intervals estimated by best model or best model set (Table 1, Supplementary Material 2). Temperature in Celsius is shown for comparison only, models were fit to inverse temperature. For clarity here, the three trophic treatments are separated into three rows of panels. Response variables were estimated once per week (for 6 weeks post bloom) in each replicate ecosystem (n = 30). For each ecosystem (shade of grey), 6 points are shown, one point for each week (symbols). Temperatures within tanks declined over time (Fig S1.1C).

In addition to the variation among ecosystems in temperature that was the main focus of our analysis, the biotic and abiotic conditions in experimental ecosystems varied over time. Temperature varied within experimental ecosystems over time (Fig S1). Overall, temperatures

458 declined between the beginning and the end of the experiment, with some variation among
459 weeks reflecting weather conditions. Phytoplankton community composition shifted over time
460 (Fig S2), but visual inspection of the species at each time point indicated no specific taxa
461 driving the changes, and there was no association between phytoplankton species composition
462 and temperature (Fig S3). Chlorophyll *a* concentration declined over time in all treatments
463 (Fig S4). A linear mixed effects model indicated that this decline was weakest in the algae-
464 only treatments (Table S6). Visual inspection of trends (Fig S4) suggests that this decline over
465 time was driven by the decline in all tanks in weeks 8 and 9, following a major rain event and
466 drop in all tank temperatures (Fig S1). When we reanalyzed the temporal trend for just weeks
467 2-7, excluding weeks 8-9, the trend in chlorophyll *a* over time approached 0 in A and AGP
468 treatments, but still persisted in AG treatments (Table S5).
469 Effects of temporal temperature variation on phytoplankton biomass within
470 ecosystems differed starkly from effects of temperature among ecosystems (Fig 6). Within
471 ecosystems, higher temperatures were associated with higher phytoplankton standing stocks
472 (Fig 6A), opposite to the trend with temperature among ecosystems. Net ecosystem oxygen
473 production (NEP) and ecosystem respiration (ER) varied with temperature within ecosystems,
474 and there is some evidence that this temperature effect interacted with both the trophic
475 structure treatment (Table 5, modelNEP8; Table 6, model ER8).
476
477
478 **Figure 6.** The effect of ecosystem temperature (T_{wj}) on A) phytoplankton biomass, B) net
479 ecosystem productivity (NEP), and C) net ecosystem respiration (ER) for three community
480 types that varied in their trophic interactions: i) algae-only (A), ii) algae + grazers (AG), and
481 iii) algae + grazers + notonectid predators (AGP). There were 10 ecosystems (*j*) in each
482 trophic treatment, and each ecosystem was sampled 8 times (once per week from weeks 2 –
483 9). Each week is indicated by a shape, and ecosystem identity within weeks are distinguished

by shades of gray. In a single model (Eqn 12), we considered effects of temperature within ecosystems over time, as well as among ecosystem variation in mean temperature (Fig 2, 5). Blue lines are fit to the 8 observations (points) from each ecosystem, and their slope indicates within-ecosystem temperature effects estimated from best models in Tables 2, 7 and 8. Black lines indicate the modelled among-ecosystem effects of temperature (Tables 1, 7, 8; Fig 2, 4). Temperature in Celsius is shown for comparison only, models were fit to inverse temperature. Temperatures within tanks declined over time (Fig S1).

Discussion

Temperature affects the metabolic rates of all organisms, and *per capita* responses to temperature of many co-occurring individuals add up to nothing less than the biological component of ecosystem scale carbon and oxygen flux. Understanding biological responses to temperature change across scales of organization (cells to the biosphere) is a major challenge in ecological research, requiring joining theoretical frameworks and empirical evidence across scales and systems. Despite much progress, there remains a gap between patterns that emerge in community-level experiments and the multi-scale theoretical framework (MTE) that links temperature dependent metabolism to larger scale patterns for temperature dependence. Here, we aimed to test the hypothesis that the effects of temperature on ecosystem processes that reflect metabolic temperature dependence are not highly sensitive to local differences in trophic structure of a community (e.g., presence or absence of a predator). This question draws upon ideas supported by the metabolic theory of ecology and community ecology theory predicting that species interactions modify the effects of temperature on community structure and function. We found that in aquatic ecosystems characterized by the presence or absence of predator-prey species interactions, temperature-dependent trophic cascades only modestly altered the effects of temperature on net ecosystem oxygen production and

consumption (NEP and ER). We found that higher average temperatures increased NEP and ER while total phytoplankton biomass declined, and all ecosystem level temperature responses were stronger than expected *per capita* temperature dependence.

Our first hypothesis was based on the expectation that our experimental systems would include trophic interactions that altered phytoplankton standing stock, and possibly interact with temperature to influence algal size distributions or other traits. We found that trophic structure did modify the effect of temperature on phytoplankton biomass, failing to reject our first hypothesis. The decline in phytoplankton standing stocks that we observed with warming across ecosystems is consistent with theoretical expectations that in closed systems with limited resources, increases in *per capita* metabolic rates with temperature could lead to declines in standing stocks [15,18,39,46]. The observed temperature dependence of phytoplankton standing stocks was highest in the communities with grazers but no predators, suggesting that temperature dependent grazing can exacerbate the temperature dependence of algal standing stocks. Overall, the temperature-dependence of phytoplankton standing stocks greatly exceeded expectations based on temperature dependence of *per capita* photosynthesis or respiration rates (Fig 3). Our hypothesis (Eqn 3) allowed for changes in phytoplankton standing stocks to be explained by direct effects of temperature on *per capita* metabolism, as well as effects of temperature on thermal traits, density or body size distributions. We suggest that change in per capita metabolic response and density were the primary components of this change. We did not observe clear shifts in the species composition of the phytoplankton assemblage with temperature; still, we do not have high resolution data on phytoplankton cell size or traits, so we cannot reject these mechanisms as contributors to the patterns we observe.

Our second hypothesis, based on recent experimental results in other freshwater and grassland systems, was that the trophic cascade would get stronger as ecosystem temperatures warmed. We found support for this hypothesis in our system, providing the first evidence that trophic cascade strength increases continuously with temperature. Prior to our study, evidence

536 of stronger trophic cascades with warming were from experiments that test two temperature
537 levels, an ambient and a simulated future scenario of ~ +3 °C [29,47,48]. We show here that
538 this pattern continues over a thermal range of 10 °C. The indirect effects of predators on
539 phytoplankton biomass appears to have been mediated by predation on the dominant grazer,
540 *Daphnia*. Predators reduced *Daphnia* density and thereby shifted grazer assemblages toward
541 the less effective copepod grazers at all temperatures. This trophic cascade, mediated by shifts
542 in grazer composition as well as total density, is a classic food web motif in freshwater
543 systems [43]. Interestingly, at warmer temperatures grazer density was lowest, yet we still
544 observed declines in biomass of phytoplankton. This pattern could reflect higher *per capita*
545 grazing by the remaining grazer individuals. Algal productivity rates are an important element
546 of trophic cascade strength [15,34], and higher NEP at warmer temperatures would contribute
547 to a stronger trophic cascade, even as grazer density declines. As with hypothesis 1, we infer
548 that the effect of temperature on the trophic cascade strength reflects not only the effect of
549 temperature on *per capita* metabolic rate but also shifts in algal traits or body sizes, or both.
550 We tested a third hypothesis that the effects of temperature on biomass and trophic
551 cascade strength would lead to distinct relationships between temperature and NEP and ER
552 for each trophic treatment type (e.g., with vs without predators). We found that the effect of
553 temperature on phytoplankton standing stock was much greater than the effects of
554 temperature on NEP or ER. For NEP and ER, there was support for a model with and
555 interaction between trophic structure and mean temperature, but for NEP a model without the
556 interaction was ranked quite highly (Table 6), and confidence intervals for the pooled
557 estimated temperature dependence do not indicate differences among trophic treatments.
558 Therefore, the strong effects of temperature on community structure (biomass, trophic
559 cascade strength) did not translate directly to net ecosystem flux rates.
560 The estimated temperature dependences of NEP and ER were greater than expected
561 based on temperature dependent *per capita*, mass normalized respiration and photosynthesis

562 metabolic rates. It is well established that temperature dependence of aerobic respiration is
563 approximately E_R = ~0.65 eV, and that this value explains the temperature dependence of
564 mass normalized ecosystem metabolism at the ecosystem scale [2-4]. The temperature
565 dependence of photosynthesis at suboptimal temperatures appears to be E_{PS} = ~0.32 eV for
566 algal systems, and this can emerge at population [39] and ecosystem scales [4] in aquatic
567 systems, suggesting E_{NEP}= ~0.32 eV, but other studies have found evidence for stronger or
568 weaker values of $E_{NEP,}$ ranging from 0 to 1.2 eV at population and ecosystem scales
569 [10,20,49]. Across our experimental temperature gradient, we observed values of E_R > ~0.65
570 eV for both NEP and ER, though confidence intervals for ER did include this value (Fig 3) for
571 algae-only ecosystems. These results led us to reject the 'first order metabolic theory'
572 hypotheses that temperature dependence of ecosystem functions scales directly with general
573 temperature dependence of metabolism. Our results further suggest that changes in species
574 interactions within communities, such as loss or gain of a predator species, could alter the
575 responses of net ecosystem fluxes to temperature changes.
576 Temperature had a stronger effect on phytoplankton standing stock than on NEP. This
577 difference in phytoplankton biomass and oxygen-flux responses to temperature could reflect
578 several processes operating at different scales of organization. First, we expect that *per capita*
579 rates of oxygen flux increase with warming, so that a given biomass of phytoplankton can be
580 more productive at warmer temperatures if resources are not limiting [4,46,50]. Patterns at the
581 ecosystem scale could deviate from expectations based on direct metabolic scaling of *per*
582 *capita* metabolism if size distributions shift toward smaller cells, as is common with warming
583 as described by the temperature size rule [23,51]. The allometric scaling of metabolic rate
584 with body size (Eqn 2) predicts greater oxygen flux for a given total biomass comprised of
585 small individuals. The distribution of thermal tolerance phenotypes may have shifted within
586 the phytoplankton communities. Three months is sufficient time for evolutionary change
587 [52]. We did not see clear evidence of shifts in species composition with temperature, and it is

588 likely that the species we collected to inoculate our ecosystems were able to tolerate our
589 experimental conditions because we collected them from a shallow lake in Vancouver in
590 which the water temperature likely tracks summertime air temperatures, therefore
591 experiencing temperatures between 19 – 30 °C.

592 In addition to the effects of temperature on *per capita* metabolism and size structure,
593 at the ecosystem scale, effective resource supply may have changed with temperature,
594 violating an implicit assumption of equations 1-4. Even though these were closed ecosystems
595 with regard to external influxes of nutrients, and they experienced the same light conditions,
596 internal nutrient processes could have varied with temperature in ways that made nutrients
597 more available in warmer ecosystems. For example, our ecosystems did not include a benthic
598 habitat that can store nutrients and organic material and slow down nutrient cycling.
599 Heterotrophic microbial processes responsible for rapid nutrient turnover would be
600 accelerated by temperature perhaps making available nutrients in warmer systems more than
601 in colder systems. Another potential, and speculative, explanation for higher productivity than
602 expected in warmer ecosystems is that some algae species are capable of biological nitrogen
603 fixation [53] and this activity is more feasible at higher temperatures. These two biological
604 processes that are themselves temperature dependent could create a resource gradient in
605 parallel with the temperature gradient [15,50], leading to higher than expected NEP at warmer
606 temperatures relative to the same ecosystem at cooler temperatures.

607 Although there was no benthic sediment in our ecosystems, algae likely colonized the
608 sides and bottom of the tanks. Benthic algae may also have contributed to NEP and ER
609 estimates in our systems [54]. We did not observe notable amounts of accumulated benthic
610 algae, but even small amounts could have contributed to total ecosystem fluxes and led to
611 covariation in total biomass with temperature. If the ratio of phytoplankton to benthic algae
612 was temperature-dependent [54], our primary producer biomass estimates may have
613 increasingly under-represented total algal biomass at higher temperatures. To be conservative,

614 we did not present mass-normalized NEP estimates because we could not normalize to any
615 benthic algal metabolic biomass. Covariation between biomass and temperature is common
616 across geographic variation in temperature [12,20,53] and therefore present in other estimates
617 of NEP across broad spatial scales when biomass cannot be estimated well.

618 Across mean ecosystem temperatures of 19 – 30 °C, we observed no sign of
619 ecosystem collapse or threshold responses to warming. Changes in community structure and
620 the increase in trophic control along the temperature gradient appear to be exponential and
621 monotonic over the 10 °C gradient (Eqn 2), suggesting that linear (or additive) models of
622 temperature effects in most warming experiments, which only test two or three temperatures,
623 may underestimate warming effects over broader thermal gradients. We observed little
624 evidence of abrupt transitions that might be expected if thermal stress responses by individual
625 phenotypes drove ecosystem scale responses. We did observe declines in grazer density with
626 warming even in the absence of predators, suggesting there were direct or indirect negative
627 effects of temperature on grazers. But we did not see clear shifts in algal species composition
628 among treatments, suggesting that no species group was exposed to temperatures above its
629 critical thermal maximum. Another challenging aspect of warming experiments at the
630 population and community scales is interpreting patterns in the context of transient dynamics.
631 Our ecosystems certainly did not reach long term states, because varying weather conditions
632 and multi-week generation times of zooplankton would have precluded that. Still, we did not
633 observe signs of transient dynamics in these communities over time such as population cycles
634 or abrupt changes.

635 In our systems, community biomass and abundance in food webs were more resistant
636 to community change with warming and longer food chains. Predators reduced zooplankton
637 density and caused a clear trophic cascade. Trophic control, and therefore any mitigating
638 effects of predators on biomass change, was weak at low temperatures and increasingly strong
639 at higher temperatures (A vs AG treatment, Fig 3). This pattern is consistent with previous

findings that ecosystem functions in systems with two (or even numbers) of trophic levels tend to be more sensitive to warming than systems with odd numbers, due to cascading effects of predation on primary producers [48]. Yet, this result contradicts theories in which dynamically responsive predators can make three-trophic-level systems dynamically less stable than shorter food chains [55]. The difference between the prediction for instability in population dynamics and stability in ecosystem function may be explained by biodiversity in our systems and functional compensation among zooplankton or phytoplankton species [56]. Additionally, in our experiment, predators were not dynamically responsive; they did not have time to reproduce during the experiment. Consequently, they represent mortality for zooplankton that may have varied with temperature effects on per capita predation rates by predators, but not demographic response. In many systems, predators are subsidized by other habitats and food sources, and their populations are not dynamically coupled to prey. In fact, this decoupling has been shown to be important in thermally stratified lakes [57]. Inferences drawn based on this experiment about how species interactions affect community and ecosystem responses are restricted to systems with dynamics in the primary producers and primary consumers, with fixed predation-related mortality imposed by a third trophic level.

The growing literature of experimental tests of how warming affects interacting species aims to reduce uncertainty in projected ecological changes associated with climate change. Warming experiments have shown a wide variety of consequences for species interactions, from shifts in community composition, strengthening top-down control, and shifts in body size [16,18,54]. We have shown that these shifts do alter the effects on the temperature dependence of net ecosystem oxygen production and consumption as modeled by the metabolic theory of ecology, but that these models may be extended to consider community level changes. By measuring community and ecosystem responses over a broad thermal gradient under controlled conditions, we have provided empirical evidence that large effects of temperature on community biomass can occur in the context of less strong effects of

temperature on net ecosystem function. This is a step toward closing the gap between patterns observed across ecosystems that appear to reflect effects of temperature on metabolic rates, and observations at intermediate scales that temperature can have large effects on the abundance of species. Taken together, these results suggest our efforts to predict community change with warming may benefit from the general metabolic scaling theory framework to understand even local-scale effects of temperature change at the community level.

Methods

Experimental Design and Set-up

We assembled freshwater food webs in 30 outdoor mesocosms (370 L tanks) at the University of British Columbia, Vancouver, Canada (49°14'52" N, 132°13'57" W). Mesocosms were filled with municipal water on June 26th, 2012, heaters were added, and filled tanks were left for one week to allow chlorine to evaporate before organisms were introduced. We experimentally manipulated temperature (10 levels) and trophic structures (algae-only (A), algae + grazer (AG), and algae + grazer + predator (AGP), Fig. 1B). There was one tank per temperature per trophic treatment; statistical power was derived from the regression design rather than replication within treatment levels (see Methods: Statistical Analysis). We monitored temperature continuously, and sampled biotic variables once per week for 9 weeks. Tanks were arranged randomly in space with regard to treatment. The spatially randomized assignment of temperature and trophic treatments eliminated systematic variation in negligible allochthonous carbon inputs.

On July 2, 2012, mesocosms were inoculated with pondwater (1L) from the UBC Pond Facility, containing living algae, collected and filtered through a 64-µm sieve to remove zooplankton and larvae. Three days later, we collected zooplankton at Trout Lake,

692 Vancouver, B.C. (49°15’23” N, 123°03’44” W), with a vertical tow net (64-µm mesh).
693 Zooplankton were mixed in buckets to homogenize species composition, were introduced to
694 mesocosm temperatures over a 12-hour gradual acclimation period to avoid stress associated
695 with an abrupt temperature change, and dead organisms were removed. Initial experimental
696 communities consisted of 25 phytoplankton taxa (Table S1), and those with zooplankton
697 included predominantly 2 zooplankton taxa (cladocerans *Daphnia* sp., and calanoid copepod
698 *Eurytemora sp.*) and, rarely, cyclopoid copepods. To ensure colonization of grazing
699 zooplankton, in addition to the random aliquot of zooplankton added to each zooplankton
700 ecosystem (all algae-grazer and algae-grazer-predator ecosystems), we added two individuals
701 of *Daphnia* sp. and ten *Eurytemora* sp. Thus each zooplankton community began with *at least*
702 12 grazing zooplankton individuals. We introduced 2 individual notonectid predators
703 (*Notonecta undulata*), collected from ponds at the UBC Pond Facility, on July 4th, 2012 to 10
704 algae-grazer-predator tanks. Notonectids generate trophic cascades by suppressing
705 zooplankton [58]. Notonectids did not reproduce during the experiment, and we replaced dead
706 notonectids during the experiment with similar-sized individuals from the same source
707 population.
708 We added 160-µg $NaNO_3$ L^{-1} and 10-µg KH_2PO_4 L^{-1} to each tank (16:1 N:P) on July
709 3rd, 2012. These quantities of nutrients represent typical deposition inputs to similar lakes
710 [59]. Water was heated with submersible aquarium heaters (50, 100, 150, 200, 250, 300, 350,
711 400, 450 Watt) to increase temperature above ambient daily temperature. Temperatures were
712 recorded hourly using Thermochron iButton dataloggers. Data loggers were suspended in the
713 middle of the tanks, approximately halfway between the surface and the bottom. Temperature
714 differences among tanks were consistent throughout the course of the experiment (Fig S1).
715 Heaters were placed at the bottom of the mesocosms. Mesocosms were covered with two
716 layers of window screen to minimize colonization by other invertebrates. Water levels were
717 maintained by natural precipitation and weekly additions to maintain volume.

718

719 **Plankton Sampling and Analysis**

720 We sampled phytoplankton, chlorophyll *a*, zooplankton, and oxygen concentrations
721 weekly until August 28th, 2012. We sampled algal assemblages in 100-mL water samples
722 collected from ~40-cm below the surface. We counted and identified cells using the Utermöhl
723 sedimentation method [60] and identified algae species or taxon level by inverted microscopy.
724 We estimated chlorophyll *a* concentration using a Trilogy fluorometer (Turner Designs).
725 Chlorophyll *a* concentration can be used as a proxy for biomass, and though the ratio between
726 chlorophyll *a* and total biomass can itself vary with temperature, size and species composition
727 [61,62], the chlorophyll *a* concentration represents biomass allocated to photosynthesis and
728 NEP, our measure of ecosystem function. We measured oxygen concentrations *in situ* using
729 YSI-85 oxygen sensor (Yellow Springs Instruments, Yellow Springs, Ohio, USA).

730 We collected zooplankton samples using a 'depth integrated zooplankton sampler'.
731 The device is a cylinder 4 cm in diameter and 60 cm in length with a cap at one end. We
732 mixed mesocosm water gently, then submerged vertically the sampler, sealed it, removed it
733 and dumped water in to a bucket. We repeated until we had removed 10 L of water, which
734 was then filtered through a 64-µm to collect zooplankton, and then the filtered water was
735 returned to mesocosms. Plankton was fixed with Lugol's iodine solution (5%). Under 10x
736 magnification, we counted and identified zooplankton to genus and measured standard length
737 for all development stages in weeks 4-8.

738

739 **Estimation of biomass and oxygen fluxes**

740 We estimated whole ecosystem oxygen fluxes using the dissolved oxygen (DO)
741 change technique [63]. Oxygen production during the daytime is the product of
742 photosynthesis minus respiration (net ecosystem production, or NEP), and oxygen depletion
743 during the night is the result of respiration (ER). We compared DO concentrations measured

over 24 hours (dawn, dusk and the following dawn). Comparison of oxygen concentrations at dawn, dusk and dawn of the following day (Eqn 5) can indicate not only the cumulative biotic NEP and ER fluxes during that time interval, but also differences in water temperature that affect oxygen concentrations in water. At standard pressure, which is appropriate for our experiment near sea level, oxygen saturation can change by approximately 1 mg/L with a change in temperature of 5 °C, described by:

$$[O_2]_E = e^{([O_2]water - [O_2]sat * \ln(T+45.93))}, \quad \text{Eqn 5}$$

where $[O_2]_{water}$ is the O_2 concentration of water, $[O_2]_{sat}$ is the concentration the water would have if it were at equilibrium with the atmosphere (390 µatm), T is temperature of the observation (°C) [64]. For the differences in temperature we observed, corrections were on the order of mean 0.0002 ± sd 0.0008 µmol O_2 / L / hr for NPP, and mean 0.0008 ± sd 0.0003 µmol O_2 / L / hr for ER. Because these values are within 25% of our total observed changes in oxygen during those periods (mean 0.003 ± sd 0.001 µmol O_2 / L / hr for NEP and mean 0.003 ± sd 0.002 µmol O_2 / L / hr for ER), we included the correction in our analyses. Overall, the conclusions based on model selection did not depend strongly on the use of the correction (results not shown).

We estimated NEP and ER by converting changes in observed O_2 (mg L^{-1}) between daytime observation times (t_{dawn}, t_{dusk}) and overnight observations (t_{dusk}, t_{dawn2}) to micromolar concentration (z = 31.25 µmol/1 mg), and correcting for changes in estimated equilibrium oxygen concentration ($[O_2]_E$) (Eqn 5) due to changes in saturation state with temperature at each time:

$$NEP = \left[\frac{([O_2]_{dusk} - [O_2]_{dawn}) - ([O_2]_{Edusk} - [O_2]_{Edawn})}{z*(t_{dusk} - t_{dawn1}}\right] \quad \text{Eqn 6a}$$

$$ER = \left[\frac{([O_2]_{dawn2} - [O_2]_{dusk}) - ([O_2]_{Edawn2} - [O_2]_{Edusk})}{z*(t_{dawn2} - t_{dusk})}\right]. \quad \text{Eqn 6b}$$

Model and hypothesis development

The expression of temperature effects on a *per capita* metabolic rate b_i – in our case, oxygen production via photosynthesis or consumption via respiration - in this model is a special case of a more complex equation that allows each species to follow a thermal performance curve (TPC), often described by a modified Sharpe-Schoolfield equation [10,20,65], in which an individual's or population's performance declines at high temperatures above some optimal temperature. We do not use this TPC model here for two reasons: we do not expect photosynthesis or respiration to exceed optimal operating temperatures in our system for most taxa based on the fact that we collected them locally from a lake and habitat type (shallow pond) near the experimental site. We model our system using equations based on Eqn 2. We believe this simpler exponential model is a suitable hypothesis for cross-system comparison in which community phenotypes or taxonomic composition may turnover along the thermal gradient [17,23]. We do not have thermal performance data for the many species in our communities that would allow fitting of thermal performance curves within communities to test an alternate approach.

We modeled M_B (Eqn 3) by including a term for trophic treatment (Z_j) in the intercept term (Eqn 3 rearranged and log transformed):

$$\ln(M_B) = ln\left(\frac{B_R}{Z_j * b_0(T_C)\langle m_B^{\alpha-1}\rangle}\right) + E_{MB}\left({}^{1}/_{kT_j} - {}^{1}/_{kT_C}\right) \quad \text{Eqn 7}$$

We derived the expression for the trophic cascade by relating algal biomass in the AGP and AG treatments:

$$\frac{M_{B.AGP}}{M_{B.AG}} = \frac{\frac{B_{R.AGP}\, e^{E_R\left({}^{1}/_{kT}{}^{-1}/_{kT_C}\right)}}{\left(b_0(T_C)\right)_{AGP}\langle m_B^{\alpha-1}\rangle_{AGP}}}{\frac{B_{R.AG}\, e^{E_R\left({}^{1}/_{kT}{}^{-1}/_{kT_C}\right)}}{\left(b_0(T_C)\right)_{AG}\langle m_B^{\alpha-1}\rangle_{AG}}} \quad \text{Eqn 8}$$

We then simplified and added temperature dependence of mass (E_m) and normalization constants (E_b). In the absence of additional information about their functional forms, we used general Arrhenius functions, but we note that other functions could be used if appropriate. Consequently, the ratio of M_B with and without predators may vary with temperature according to the relative temperature dependences of thermal traits and size distributions:

$$\frac{M_{B.AGP}}{M_{B.AG}} \propto \frac{b_0(T_C)_{AG} e^{-E_{b.ag}/kT} \langle m_B^{\alpha-1} \rangle_{AG} e^{-E_{m.ag}/kT}}{b_0(T_C)_{AGP} e^{-E_{b.agp}/kT} \langle m_B^{\alpha-1} \rangle_{AGP} e^{-E_{m.agp}/kT}} \quad \text{Eqn 9}$$

and the strength of the trophic cascade may therefore be expected to decline with a temperature dependence that reflects the temperature dependences of mass and normalized performance for each trophic treatment:

$$ln\left(\frac{M_{B.AGP}}{M_{B.AG}}\right) \propto \frac{\ln(b_0(T_C)_{AG}) + ln\left(\langle m_B^{\alpha-1} \rangle_{AG}\right) - \frac{E_{b.ag} - E_{m.ag}}{kT}}{\ln(b_0(T_C)_{AGP}) + ln\left(\langle m_B^{\alpha-1} \rangle_{AGP}\right) - \frac{E_{b.agp} - E_{m.agp}}{kT}} \quad \text{Eqn 10}$$

We modeled zooplankton density (N / L) as a function of mean weekly ecosystem temperature T_{wj} and ecosystem trophic structure Z_j, with ecosystem identity as a random effect.

Statistical Analysis

We tested our hypotheses about whether effects of temperature on metabolism are modified at the ecosystem level by species interactions using a regression experimental design involving 30 independent ecosystems (Fig S1). We maintained ecosystems at distinct temperatures in a regression design with mean ecosystem temperatures T_{wj} ranging from 19.7 ($\pm$ 3.15) °C to 26.1 ($\pm$ 3.59) °C (Fig S1). The regression design allowed us to estimate slopes (e.g., E_R, Eqn 2) of response variables along a continuous temperature gradient for different trophic structures (A, AG, AGP) by log-transforming equation 2 and fitting linear models to log

816 transformed response variables the continuous temperature gradient. We chose the regression
817 design, though unreplicated within temperature levels, because it allowed us to compare
818 activation energies (E_R, Eqn 2) fitted over a broad range of temperatures; an important test of
819 thermal responses that is not possible with designs with only 2 or even three temperature
820 levels. Regression designs, even without replication within levels, gain statistical power from
821 the range of x-levels tested [66,67].

822 We used a mixed effects model (lme function in the nlme package of R) to examine
823 the main and interactive effects of temperature (a continuous fixed factor) and trophic
824 structure (a categorical fixed factor) on net ecosystem oxygen production (NEP), net
825 ecosystem oxygen consumption (ER), and chlorophyll *a* concentration with a random
826 intercept for individual ecosystems. We used a within-subject mean centering approach to
827 distinguish temperature effects into those associated with an ecosystem's average temperature
828 (T_j) over the entire experimental period (a 'between-ecosystem' effect) from effects variation
829 in temperature over time (T_{wj}) (a 'within-ecosystem temperature' effect) [68]. The response
830 variable (Y) for each ecosystem *j* in week *w* was modelled as a continuous response to
831 variation in inverted ecosystem temperature ($1/kT_{wj}$) and trophic treatment (Z_j):

832 $$\ln(Y_{wj})$$

833 $$= \beta_{0.j(w)} + \beta_1\left({}^1\!/_{kT_{wj}} - {}^1\!/_{k\bar{T}_j}\right) + \beta_2\left({}^1\!/_{k\bar{T}_j}\right) + \beta_3\left({}^1\!/_{kT_{wj}} - {}^1\!/_{k\bar{T}_j}\right)\left({}^1\!/_{k\bar{T}_j}\right) + \beta_4 Z_j$$

834 $$+ \beta_5 Z_j\left({}^1\!/_{k\bar{T}_j}\right) + \beta_6 Z_j\left({}^1\!/_{k\bar{T}_j} - {}^1\!/_{k\bar{T}_j}\right) + u_j + e_{wj}$$

835 Eqn 11

836 where $\beta_{0.j(i)}$ represents an intercept allowed to vary randomly among ecosystems. The terms
837 in the full model (Eqn 11) are: the between-ecosystem effect of temperature (β_2), estimated as
838 the slope of $\ln(Y_{wj})$ on the mean temperature over all weeks for ecosystem *j*, expressed as

inverse temperature $\left({}^{1}/_{k\bar{T}_j}\right)$; the within-ecosystem ($\beta_1$) effect of temperature variation over time estimated as the slope of $\ln(Y_{wj})$ *vs* centered weekly temperature $\left({}^{1}/_{k\bar{T}_{wj}}\right) - \left({}^{1}/_{k\bar{T}_j}\right)$; interaction ($\beta_3$) between within-ecosystem temporal variation in temperature and the experimental temperature treatment; trophic species interactions (β_4), and interactions between species interactions and overall mean (β_5) and weekly temperature (β_6).

To test our hypothesis that species interactions modify temperature dependence (E_R, Eqn 2) of response variables (Y), we compared models with and without trophic level terms (β_4) and interactions between Z_j and temperature (β_5, β_6). We also tested models without temperature terms for within-system variation (β_4). In total, the model set included 9 models (Table 1). Response variables were ln-transformed prior to analyses to achieve normal distributions and to linearize temperature effects for analysis and to fit E_R values from Eqn 2. When modelling, we centered temperature treatment ($1/kT_j$) on the grand mean of all temperatures observations $\bar{T}$ (not shown in Eqn 11) to reduce correlations between slope and intercept terms [69].

To test the effect of temperature on trophic cascade strength, we used the following statistical model:

$$\ln(TC_{ij}) = \beta_{0.p(w)} + \beta_1 * \left({}^{1}/_{kT_{wp}} - {}^{1}/_{k\bar{T}_p}\right) + \beta_2 * w + \beta_3 * \left({}^{1}/_{kT_{wp}} - {}^{1}/_{k\bar{T}_p}\right) * w + u_p + e_{wp} \quad \text{Eqn 12,}$$

in which the effect of temperature on trophic cascade strength in each temperature treatment j was modeled for each week w and for the temperature of the tanks, with random effects u_j were assigned for each power treatment (p).

We ranked models using Akaike's Information Criterion weights (using the MuMin package in R), adjusted for small sample sizes (AIC_C). When two or more models were considered comparable or equivalent ($\delta AIC_C < 2$) we reported all models meeting this

criterion and report averaged coefficients. We estimated the realized temperature dependence of our response variables (slopes) and intercepts for among-ecosystem responses to temperatures by first rearranging Eqn 12 to group coefficients by temperature term.

$$\ln(Y_{jw}) = \beta_{0.j(w)} + \left(\beta_1 + \beta_3 * \left({}^{1}/_{k\bar{T}_j}\right) + \beta_6 * Z_j\right)\left({}^{1}/_{kT_{wj}} - {}^{1}/_{k\bar{T}_j}\right) + \left(\beta_2 + \beta_5 * Z_j\right) * \left({}^{1}/_{k\bar{T}_j}\right) + \beta_4 * Z_j + u_j + e_{wj}. \quad \text{Eqn 13}$$

The pooled coefficients $(\beta_2 + \beta_5 * Z_j)$ include the temperature dependence of ecosystem responses plus any variation with trophic structure, and gives the slope of lines plotted in figures 2A and 5. The intercept is set by $\beta_{0.j(w)}$+$\beta_4 * Z_j$, and the remaining coefficient gives within-tank variation as plotted in Figure 6. We estimated confidence intervals for composite terms following [70]. We used R statistical software (R v. 1.0.44 R Developmental Core Team 2006). Our models controlled for the effect of temperature variation over time on ecosystem fluxes and biomass within systems.

We determined the effects of temperature and predator presence on zooplankton abundance data using generalized linear mixed effects models with tank as a random effect modeled on a negative binomial regression distribution to account overdispersed Poisson distributed count data (using the glmmADMB package in R).

Acknowledgments

We thank S. Pawar and his lab group, J. Bernhardt, R. Elahi and P. Thompson for comments and feedback that improved this manuscript, W. Cheung and F. Ratcliffe for sampling assistance and D. Song for assistance during zooplankton identification. UBC Mobility Funds to JG, NSERC Discovery Grant to MO.

References

1. Gillooly JF, Enquist BJ, Brown JH, West GB, Savage V, Charnov EL. Effects of Size and Temperature on Metabolic Rate. Science. 2001;293: 2248–2251.

2. Allen AP, Gillooly JF, Brown JH. Linking the global carbon cycle to individual metabolism. Funct Ecol. Wiley/Blackwell (10.1111); 2005;19: 202–213. doi:10.1111/j.1365-2435.2005.00952.x

3. Yvon-Durocher G, Caffrey JM, Cescatti A, Dossena M, del Giorgio P, Gasol JM, et al. Reconciling the temperature dependence of respiration across timescales and ecosystem types. Nature. Nature Publishing Group; 2012;487: 472–476. doi:10.1038/nature11205

4. Lopez-Urrutia Á. Scaling the metabolic balance of the oceans. Proc Natl Acad Sci USA. 2006;: 1–6.

5. Louca S, Polz MF, Mazel F, Albright MBN, Huber JA, O'Connor MI, et al. Function and functional redundancy in microbial systems. Nat ecol evol. Nature Publishing Group; 2018;2: 936–9. doi:10.1038/s41559-018-0519-1

6. Yvon-Durocher G, Dossena M, Trimmer M, Woodward G, Allen AP. Temperature and the biogeography of algal stoichiometry. Global Ecology and Biogeography. 2015;24: 562–570. doi:10.1111/geb.12280

7. Lopez-Urrutia Á. The metabolic theory of ecology and algal bloom formation. Limnol Oceanogr. 2008;53: 2046–2047.

8. Cheung WWL, Watson R, Pauly D. Signature of ocean warming in global fisheries catch. Nature. Nature Publishing Group; 2013;497: 365–368. doi:10.1038/nature12156

9. Anderson-Teixeira KJ, Vitousek PM, Brown JH. Amplified temperature dependence in ecosystems developing on the lava flows of Mauna Loa, Hawai'i. Proc Natl Acad Sci USA. 2008;105: 228–233. doi:10.1073/pnas.0710214104

10. Barneche DR, Kulbicki M, Floeter SR, Friedlander AM, Maina J, Allen AP. Scaling metabolism from individuals to reef-fish communities at broad spatial scales. Worm B, editor. Ecology Letters. 2014;17: 1067–1076. doi:10.1111/ele.12309

11. Kerkhoff AJ, Enquist BJ, Elser JJ, Fagan WF. Plant allometry, stoichiometry and the temperature-dependence of primary productivity. Global Ecology and Biogeography. 2005;14: 585–598. doi:10.1111/j.1466-822X.2005.00187.x

12. Michaletz ST, Cheng D, Kerkhoff AJ, Enquist BJ. Convergence of terrestrial plant production across global climate gradients. Nature. Nature Publishing Group; 2014;39: 1–13. doi:10.1038/nature13470

13. Uszko W, Diehl S, Englund G, Amarasekare P. Effects of warming on predator-prey interactions - a resource-based approach and a theoretical synthesis. Brose U, editor. Ecology Letters. 2017;20: 513–523. doi:10.1111/ele.12755

14. Rall BC, Vucic-Pestic O, Ehnes RB, Emmerson M, Brose U. Temperature, predator-prey interaction strength and population stability. Global Change Biol. 2009;16: 2145–2157. doi:10.1111/j.1365-2486.2009.02124.x

15. Gilbert B, Tunney TD, McCann KS, DeLong JP, Vasseur DA, Savage V, et al. A bioenergetic framework for the temperature dependence of trophic interactions. Wootton T, editor. Ecology Letters. 2014;17: 902–914. doi:10.1111/ele.12307

16. Shurin JB, Clasen JL, Greig HS, Kratina P, Thompson PL. Warming shifts top-down and bottom-up control of pond food web structure and function. Philos Trans R Soc Lond, B, Biol Sci. 2012;367: 3008–3017. doi:10.1098/rstb.2012.0243

17. O'Connor MI, Piehler MF, Leech DM, Anton A, Bruno JF. Warming and Resource Availability Shift Food Web Structure and Metabolism. Loreau M, editor. 2009;7: e1000178. doi:10.1371/journal.pbio.1000178

18. O'Gorman EJ, Zhao L, Pichler DE, Adams G, Friberg N, Rall BC, et al. Unexpected changes in community size structure in a natural warming experiment. 2017;7: 659–663. doi:10.1038/nclimate3368

19. Brauer VS, de Jonge VN, Buma AGJ, Weissing FJ. Does universal temperature dependence apply to communities? An experimental test using natural marine plankton assemblages. Oikos. 2009;118: 1102–1108. doi:10.1111/j.1600-0706.2009.17371.x

20. Padfield D, Lowe C, Buckling A, Ffrench-Constant R, Student Research Team, Jennings S, et al. Metabolic compensation constrains the temperature dependence of gross primary production. Ecology Letters. 2017;20: 1250–1260. doi:10.1111/ele.12820

21. Atkinson D. Temperature and Organism Size - A Biological Law for Ectotherms? Advances in Ecological Research. 1994;25: 1–58.

22. DeLong JP. Experimental demonstration of a "rate–size" trade-off governing body size optimization. Evolutionary Ecology Research. 2012;14: 343–352.

23. Forster J, Hirst AG, Atkinson D. Warming-induced reductions in body size are greater in aquatic than terrestrial species. Proc Natl Acad Sci USA. 2016;109: 19310–19314. doi:10.1073/pnas.1210460109

24. Englund G, Öhlund G, Hein CL, Diehl S. Temperature dependence of the functional response. Ecology Letters. 2011;14: 914–921. doi:10.1111/j.1461-0248.2011.01661.x

25. Siegle MR, Taylor EB, O'Connor MI. Prior heat accumulation reduces survival during subsequent experimental heat waves. Journal of Experimental Marine Biology and Ecology. Elsevier; 2018;501: 109–117. doi:10.1016/j.jembe.2018.01.012

26. Dell AI, Pawar S, Savage VM. Systematic Variation in the Temperature Dependence of Physiological and Ecological Traits. 2011 May pp. 1–64.

27. Sentis A, Ramon-Portugal F, Brodeur J, Hemptinne J-L. The smell of change: warming affects species interactions mediated by chemical information. Global Change Biol. 2015;21: 3586–3594. doi:10.1111/gcb.12932

28. Sentis A, Binzer A, Boukal DS. Temperature-size responses alter food chain persistence across environmental gradients. Vasseur D, editor. Ecology Letters. 2017;20: 852–862. doi:10.1111/ele.12779

29. Kratina P, Greig HS, Thompson PL, Carvalho-Pereira TSA, Shurin JB. Warming modifies trophic cascades and eutrophication in experimental freshwater communities. Ecology. John Wiley & Sons, Ltd; 2012;93: 1421–1430. doi:10.1890/11-1595.1

30. Atwood TB, Hammill E, Greig HS, Kratina P, Shurin JB, Srivastava DS, et al. Predator-induced reduction of freshwater carbon dioxide emissions. Nature Geoscience. Nature Publishing Group; 2013;6: 191–194. doi:10.1038/ngeo1734

31. Barton BT, Beckerman AP, Schmitz OJ. Climate warming strengthens indirect interactions in an old-field food web. Ecology. 2009;90: 2346–2351. doi:10.1890/08-2254.1

32. Schindler DE, Carpenter SR, Cole JJ, Kitchell JF, Pace ML. Influence of food web structure on carbon exchange between lakes and the atmosphere. Science. 1997;277: 248–251.

33. Polis GA, Sears ALW, Huxel GR, Strong DR, Maron J. When is a trophic cascade a trophic cascade? Trends in Ecology & Evolution. 2000;15: 473–475.

34. DeLong JP, Gilbert B, Shurin JB, Savage VM, Barton BT, Clements CF, et al. The Body Size Dependence of Trophic Cascades. The American Naturalist. 2015;185: 354–366. doi:10.1086/679735

35. Estes JA, Terborgh J, Brashares JS, Power ME, Berger J, Bond WJ, et al. Trophic Downgrading of Planet Earth. Science. 2011;333: 301–306. doi:10.1126/science.1205106

36. Atwood TB, Hammill E, Kratina P, Greig HS, Shurin JB, Richardson JS. Warming alters food web-driven changes in the CO2 flux of experimental pond ecosystems. Biology Letters. 2015;11: 20150785. doi:10.1098/rsbl.2015.0785

37. Brown JH, Gillooly JF, Allen AP, Savage VM, West GB. Toward a Metabolic Theory of Ecology. Ecology. 2004;85: 1771–1789.

38. Price CA, Weitz JS, Savage VM, Stegen J, Clarke A, Coomes DA, et al. Testing the metabolic theory of ecology. Ecology Letters. 2012;: 1–10. doi:10.1111/j.1461-0248.2012.01860.x

39. Bernhardt JR, Sunday JM, O'Connor MI. Metabolic Theory and the Temperature-Size Rule Explain the Temperature Dependence of Population Carrying Capacity. The American Naturalist. 2018;192: 687–697. doi:10.1086/700114

40. Katechakis A, Stibor H, Sommer U, Hansen T. Changes in the phytoplankton community and microbial food web of Blanes Bay (Catalan Sea, NW Mediterranean) under prolonged grazing pressure by doliolids (Tunicata), cladocerans or copepods (Crustacea). Mar Ecol Prog Ser. 2002;234: 55–69.

41. Osmond MM, Barbour MA, Bernhardt JR, Pennell MW, Sunday JM, O'Connor MI. Warming-Induced Changes to Body Size Stabilize Consumer-Resource Dynamics. 2017;189: 718–725. doi:10.1086/691387

42. Shurin JB, Borer E, Seabloom E, Anderson K, Blanchette C, Broitman B, et al. A cross-ecosystem comparison of trophic cascades. 2002;6: 785–791.

43. Brooks JL, Dodson SI. Predation, body size, and composition of plankton. Science. 1965;150: 28–35.

44. Barton BT, Beckerman AP, Schmitz OJ. Climate warming strengthens indirect interactions in an old-field food web. Ecology. 2009;90: 2346–2351.

45. Svensson F, Karlsson E, Gårdmark A, Olsson J, Adill A, Zie J, et al. In situ warming strengthens trophic cascades in a coastal food web. Oikos. 2017;126: 1150–1161. doi:10.1111/oik.03773

46. O'Connor MI, Gilbert B, Brown CJ. Theoretical Predictions for How Temperature Affects the Dynamics of Interacting Herbivores and Plants. 2011;178: 626–638. doi:10.1086/662171

47. Mckee D, Hatton K, Eaton JW, Atkinson D, Atherton A, Harvey I, et al. Effects of simulated climate warming on macrophytes in freshwater microcosm communities. Aquat Bot. 2002;74: 71–83. doi:10.1016/S0304-3770(02)00048-7

48. Hansson L-A, Nicolle A, Graneli W, Hallgren P, Kritzberg E, Persson A, et al. Food-chain length alters community responses to global change in aquatic systems. 2013;3: 228–233. doi:10.1038/nclimate1689

49. Sal S, Garcia-Carreras B, Sheppard R, Rizzuto M, Etard A, Yvon-Durocher G, et al. Metabolic mismatches and compensation in the thermal dependence of daily carbon flux in plants. 2017;: 1–12.

50. Cross WF, Hood JM, Benstead JP, Huryn AD, Nelson D. Interactions between temperature and nutrients across levels of ecological organization. Global Change Biol. 3rd ed. 2015;21: 1025–1040. doi:10.1111/gcb.12809

51. Garzke J, Hansen T, Ismar S, Sommer U. Combined Effects of Ocean Warming and Acidification on Copepod Abundance, Body Size and Fatty Acid Content. PLoS ONE. 2016;11: e0155952.

52. Padfield D, Yvon-Durocher G, Buckling A, Jennings S, Yvon-Durocher G. Rapid evolution of metabolic traits explains thermal adaptation in phytoplankton. Hillebrand H, editor. Ecology Letters. 2015;19: 133–142. doi:10.1111/ele.12545

53. Welter JR, Benstead JP, Cross WF, Hood JM, Huryn AD, Johnson PW, et al. Does N2 fixation amplify the temperature dependence of ecosystem metabolism? Ecology. 2015;96: 603–610.

54. Dossena M, Yvon-Durocher G, Grey J, Montoya JM, Perkins DM, Trimmer M, et al. Warming alters community size structure and ecosystem functioning. Proc R Soc B. 2012;279: 3011–3019. doi:10.1098/rspb.2012.0394

55. Hastings A, Powell T. Chaos in a three-species food chain. Ecology. 1991;72: 896–903.

56. Loreau M, Mouquet N, Gonzalez A. Biodiversity as spatial insurance in heterogeneous landscapes. Proc Natl Acad Sci USA. 2003;100: 12765–12770.

57. Tunney TD, McCann KS, Lester NP, Shuter BJ. Effects of differential habitat warming on complex communities. Proc Natl Acad Sci USA. 2014;: 1–6. doi:10.1073/pnas.1319618111

58. McArdle BH, Lawton JH. Effects of prey-size and predator-instar on the predation of Daphnia by Notonecta. Ecol Entomol. Blackwell Publishing Ltd; 1979;4: 267–275. doi:10.1111/j.1365-2311.1979.tb00584.x

59. Carpenter SR, Caraco NF, Correll DL, Howarth RW, Sharpley AN, Smith VH. Nonpoint pollution of surface waters with phosphorus and nitrogen. Ecological Applications. 1998;8: 559–568.

60. Utermöhl H. Zur Vervollkommnung der quantitativen Phytoplankton-Methodik. Mitteilungen Internationale Vereiningung fuer Theoretische und Angewandte Limnologie. 1958;9: 1–38.

61. Raven JA, Geider RJ. Temperature and algal growth. New Phytol. 1988;110: 441–461.

62. Geider RJ, MacIntyre HL, Kana TM. Dynamic model of phytoplankton growth and acclimation: responses. Mar Ecol Prog Ser. 1997;148: 287–200.

63. Marzolf ER, Mulholland PJ, Steinman AD. Improvements to the Diurnal Upstream–Downstream Dissolved Oxygen Change Technique for Determining Whole-Stream Metabolism in Small Streams. Can J Fish Aquat Sci. NRC Research Press; 1994;51: 1591–1599. doi:10.1139/f94-158

64. Moore ML. NALMS management guide for lakes and reservoirs. North American Lake Management Society. North American Lake Management Society. 1989.

65. Schoolfield RM. Non-linear regression of biological temperature-dependent rate models based on absolute reaction-rate theory. Journal of Theoretical Biology. 1981;88: 719–731.

66. Cottingham KL, Cottingham KL, Lennon JT, Lennon JT, Brown BL, Brown BL. Knowing when to draw the line: designing more informative ecological experiments. Frontiers in Ecology and the Environment. Ecological Society of America; 2005;3: 145–152. doi:10.1890/1540-9295(2005)003[0145:KWTDTL]2.0.CO;2

67. Gotelli NJ, Ellison AM. Chapter 7: A Bestiary of Experimental & Sampling Designs. A Primer of Ecological Statistics. 2nd ed. Sunderland, Massachusetts, USA: Sinauer Associates Incorporated; 2013. p. 614. Available: http://www.worldcat.org/title/primer-of-ecological-statistics/oclc/814529364

68. van de Pol M, Wright J. A simple method for distinguishing within- versus between-subject effects using mixed models. Animal Behaviour. Animal Behaviour; 2009;77: 753–758. doi:10.1016/j.anbehav.2008.11.006

69. O'Connor M, Bruno JF, Gaines SD, Halpern BS, Lester S, Kinlan BP, et al. Temperature control of larval dispersal and the implications for marine ecology, evolution, and conservation. Proc Natl Acad Sci USA. 2007;104: 1266–1271.

70. Figueiras A, Domenech-Massons JM, Cadarso C. Regression models: calculating the confidence interval of effects in the presence of interactions. Stat Med. 1998;17: 2099–2105. Available: http://eutils.ncbi.nlm.nih.gov/entrez/eutils/elink.fcgi?dbfrom=pubmed&id=9789916&retmode=ref&cmd=prlinks