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The effect of temperature and predation on performance in monoculture and in competition in three Daphniidae differing in body size

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Abstract

Zooplankton body size shows a strong association with temperature, competition, and predation. Global warming affects all three drivers of body size and is thus expected to lead to substantial changes in zooplankton community composition and body size distributions. To disentangle the isolated and joint effect of temperature, competition, and fish predation on species biomass and community composition in zooplankton, we monitored population biomasses of three Daphniidae species that differ in body size (*Daphnia magna*, *Daphnia pulex*, and *Ceriodaphnia reticulata*) for 20 days, manipulating competition (monoculture, pairwise trials, and three-species communities), temperature (20°C, 24°C, and 28°C) and presence or absence of fish predation. In the absence of predation, *D. magna* dominated in all competition experiments, even at high temperatures. *D. magna* went extinct, however, in the predation treatments at 24°C and 28°C. *D. pulex* outcompeted *C. reticulata* and was negatively affected by predation and high temperature. *C. reticulata* did not reduce biomass at high temperatures and was negatively affected by all competition trials, but was positively affected by predation. Our results indicate that the two larger-bodied species are more negatively affected by the combination of temperature and predation than the smallest species. While higher temperatures reduced the biomass of the larger-bodied species, it did not fundamentally change their ability to dominate over the smallest species in competition. The combined effect of warming and predation changed community composition more fundamentally, resulting in the dominance of small-bodied species. This can have important ecosystem-wide impacts, such as the transition to turbid, algae-dominated systems.

Ongoing climate warming has by now affected all levels of biological organization (Parmesan 2006; Scheffers et al. 2016). While aquatic ecosystems and their biota have been intensively studied in the context of climate change, it has remained a challenge to accurately predict and verify the effects of global warming on freshwater communities and ecosystems services (Rosset et al. 2010; Jeppesen et al. 2014). A key reason for this is the complexity of processes that interact in determining the outcome of such a fundamental change as climate warming (Urban et al. 2016). This complexity is not only related to how individuals and populations can respond to environmental change via phenotypic plasticity and evolutionary trait change

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Additional Supporting Information may be found in the online version of this article.

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(Van Doorslaer et al. 2009; Brans et al. 2017; De Meester et al. 2018) or migration (Norberg et al. 2012) but also strongly depends on species interactions and how these influence the performance of focal species (Urban et al. 2016).

Warming effects at the level of an organism alter metabolic rates and thus influence growth, reproduction, respiration, and feeding rates (Lear et al. 2020; Bomfim et al. 2022). Experimental and biogeographic studies have shown that warming influences the body size of organisms (Forster et al. 2012; Havens et al. 2014), where smaller body sizes are expected at high temperatures (Moore et al. 1996; Daufresne et al. 2009). The temperature size rule predicts that ectotherms mature at smaller body sizes with increasing temperature in response to greater metabolic demands (Atkinson 1994; Verberk et al. 2021). In addition, oxygen becomes more limiting in aquatic systems at higher temperatures, which can also select for smaller body sizes, as this allows more efficient transport of oxygen to all effector organs (Atkinson et al. 2001; Verberk et al. 2021). Species can also, however, adapt to warming through physiological and biochemical changes independent of body size changes, such as increased production of hemoglobin and heat shock proteins (Gerke et al. 2011; Zeis et al. 2013).

The metabolic changes driven by global warming are also expected to intensify some ecological relationships in aquatic ecosystems, such as competition (Moore et al. 1996) and predation (Thakur et al. 2018). Herbivorous cladocerans, for example, and especially members of the Daphniidae family, share similar ecological niches and compete strongly for food resources (Hu and Tessier 1995; Adamczuk 2010). In general, large-bodied Daphniidae species suppress small-bodied species because they attain higher ingestion rates as a result of a wider prey size spectrum, generalist diet, and higher energy efficiency (Gliwicz 1980; Jiang et al. 2014). In a warmer environment, where organisms have higher consumption rates linked to higher metabolic demands (Forster et al. 2011) and food quality may be insufficient (Visser et al. 2016), large-bodied cladocerans might be better competitors as they are more efficient filter feeders (Gliwicz 1980). Yet, they might be suppressed by small-bodied cladocerans, as high temperature favors small ectotherms (Moore et al. 1996). Likewise, the increase in reproduction rates and organism densities at high temperatures (Kooijman 2010) can lead to increased competition (Forster et al. 2011). The outcome of competition under climate warming will be determined by the degree to which the different species have a low food threshold through an optimal resource allocation in reproduction and survival at high temperatures (Glazier and Calow 1992).

Besides competition, predation is also influenced by temperature, with the potential to alter the structure of the zooplankton community via body size. Warming leads to higher predation pressure by fish on zooplankton due to higher metabolic demands and increased consumption rates of fish (Meerhoff et al. 2007). Visual predation by fish is biased towards large-bodied species and is since long recognized as one of the key structuring processes in zooplankton communities (Brooks and Dodson 1965; Lampert and Sommer 2011). The prediction that both temperature and fish predation should shift the size distribution of zooplankton to smaller species and individuals is supported by experimental studies (Hart and Bychek 2011; He et al. 2018) and the observation that in lowland tropics the zooplankton is dominated by small species (Iglesias et al. 2011; Ersoy et al. 2017).

Warming, competition, and predation interact to determine the structure of zooplankton communities and their body size distribution. However, the direct and indirect effects of increased metabolic demands lead to conflicting predictions on whether body sizes should increase or decrease under competition and predation in response to climate warming. Predation by fish has a direct effect on the shift of zooplankton body size distribution to smaller individuals but also reduces competition by selectively feeding on large-bodied zooplankton, and this reduction in competition may also affect zooplankton body size distributions (Chase et al. 2002; Iglesias et al. 2011). Temperature can favor small-bodied species (Atkinson 1994; Verberk et al. 2021), but indirectly can also increase both competition and predation (Moore et al. 1996). Whereas large-bodied cladocerans are better competitors, they can be more affected by temperature (Verberk et al. 2021). These responses to climate warming are, thus, expected to result in profound shifts in the size structure of populations and communities, and the structure of food webs (Thébault and Loreau 2003).

To disentangle the effects of warming, fish predation, and competition on zooplankton, we carried out a microcosm experiment testing for the isolated and joint effect of these three factors on population biomass and species composition using three cladocerans of the Daphniidae family that differ in body size: the large-bodied *D. magna* (Straus, 1820), the medium-sized *D. pulex* (Leydig, 1860), and the smaller-bodied *C. reticulata* (Jurine, 1820). We assessed whether and to what extent the direct effect of temperature alone can alter the relative dominance of large- vs. small-bodied cladocerans in competitive contexts, or whether fish predation pressure is needed to enforce such a change. We expected that (1) warming would negatively affect the population biomass of all species in monoculture, but with stronger effects on the larger-bodied species compared to smaller species, (2) warming would also reduce species-specific biomass in competition trials and would alter the relative dominance of large- vs. small-bodied cladocerans toward the dominance of small-bodied cladocerans, (3) predation would have a strong negative impact on biomass, again affecting larger-bodied species more than smaller-bodied, and the effect would be intensified at high temperatures, finally, (4) predation pressure on large-bodied species would favor small-bodied species in competition trials, changing community composition toward the dominance of small-bodied species.

Materials and methods

Study organisms and culture conditions

D. magna, *D. pulex*, and *C. reticulata* were sampled from ponds in the region of Leuven and Ghent, Belgium over several years (for information confirming the clear differences in body size among the three species, see Data S1). Individual females collected from the field, were isolated and cultivated as clonal lineages. From all clonal lineages present in our stock collection, we randomly chose 10 clones per species and cultured them in mixed populations for ~ three months under standardized conditions in one-liter glass jars filled with dechlorinated (24h aged) tap water and kept under a 14 : 10h L : D photoperiod (Fig. 1). Twenty multiclonal cultures per species were kept at each of the three experimental temperatures (20°C, 24°C, and 28°C). We chose these specific temperatures as the first two reflect the average summer temperature in Belgium in rural (20°C) and urban ponds (24°C) (Brans et al. 2018), whereas 28°C was chosen based on a + 4°C climate forecast (IPCC 2014). Half of the culture medium was renewed twice a week. All cultures were fed with a 1 : 1 (biomass) mixture of two different batch-cultured algae species, *Chlorella vulgaris* (Beyerinck) and *Acutodesmus obliquus* (Turpin-Kützing) at a total concentration of 1 mg C^{-1} L^{-1}. We set up a length-weight regression to estimate individual biomasses and set up a life table experiment for each species at the three experimental temperatures. A more in-depth description of the methods used in length–weight regression and life table as well as the resulting values are given in the Data S1.

We chose Pumpkinseed sunfish, *Lepomis gibbosus* (Linnaeus 1758) with body size varying between 8 and 10 cm as the predator because they occur in Belgian ponds (as an exotic but locally abundant species) and are very stress-resistant and able to cope with our experimental temperatures (Power and Todd 1976). The fish were maintained in aquaria with air pump, kept at the three experimental temperatures (20°C, 24°C, and 28°C), and fed with Chironomidae larvae. Throughout our study, we followed the protocol stipulated and approved by the Ethical Commission from the Katholieke Universiteit Leuven under registration P006/2019 (LA1210204).

Microcosm experiment

We carried out an indoor microcosm experiment in which we included monocultures of each species as well as competition trials with all three pairwise combinations of two species and the three species combination (Fig. 1). We carried out the competition experiment at three experimental temperatures (20°C, 24°C, and 28°C) and both in the absence and presence of fish predation, except for the monocultures, which were only studied in the absence of fish predation. All experimental units were performed in triplicate leading to the following design: [3 monocultures + (4 competition combinations (three pairwise, one full combination) × 2 fish treatments (presence/absence))] × 3 temperatures (20°C, 24°C, and 28°C) × 3 replicates = 99 experimental units (Fig. 1).

Each microcosm consisted of a 6 liter aquarium filled with 5 liter of dechlorinated tap water. All microcosms were placed in a temperature-controlled water bath (20°C, 24°C and 28°C ± 1°C) at 14 : 10h L : D photoperiod and contained artificial plants (three plastic *Elodea canadensis* mimics of 20 cm high) providing refuge from predation. Half of the experimental medium (dechlorinated tap water) was renewed twice a week. Food was replenished to 1 mg C^{-1} L^{-1} of a 1 : 1 mixture of *C. vulgaris* and *A. obliquus* every two days. The experiment started by inoculating adults randomly chosen from bottles with 10 different clones per species kept at the experimental temperature (i.e., preconditioned to that temperature). The number of individuals of each species in each aquarium was standardized to equal biomass, taking into account the differences in biomass of adult females of each species (*D. magna* = 28.6 μg DW ind^{-1}; *D. pulex* = 8.1 μg DW ind^{-1}, and *C. reticulata* = 3.2 μg DW ind^{-1}; see Data S1). In each aquarium, we inoculated a total number of individuals that corresponds to the biomass of 10 adult *D. magna* (i.e., 286 μg DW). The resulting number of individuals of each species inoculated in the aquaria is given in Fig. 1 and Table S2.

The competition experiment ran for 20 days. The predation treatment involved a repeated and timed exposure to an actively hunting fish. We carefully put one fish (randomly selected out of a group of 15 fish that were kept in a 50 liter aquarium at the corresponding temperature, i.e., 20°C, 24°C, or 28°C, total number of fish = 45) for exactly 10 min (measured with a chronometer) in each microcosm of the predation treatment on day 10, 15, and 20. Pilot experiments had established that a substantial proportion of the cladocerans were eaten during these 10 min time-periods.

All communities and populations were completely harvested at the end of the experiment (day 20; water volume = 5 liter). Samples were fixed with sugar formaldehyde (4%). All samples were fully counted and categorized into size classes (juveniles, adults, adults with eggs). Densities (individuals L^{-1}) were transformed into biomass (μg DW L^{-1}) by multiplying the number of individuals of each size class by the average estimated dry weight per individual of that size class.

Statistical analyses

To test our hypotheses, we performed different ANOVAs using species-specific biomass as a response variable. Because our experimental design did not contain all full factorial treatment combinations (i.e., monocultures treatments were not tested for fish predation), we split up the analysis into two parts: one testing the interaction between temperature and competition in the absence of fish and a second one testing the three-way interaction between temperature, competition and fish presence for all competition trials. In all analyses outlined below model assumptions were checked and were met unless stated otherwise. We visually checked for deviations in model residuals normality using qqplots and histograms (Zuur et al. 2010). Significant main or interaction effects were followed by Tukey post hoc comparisons to assess which treatments significantly differ from each other ("emmeans" R package; Lenth et al. 2019).

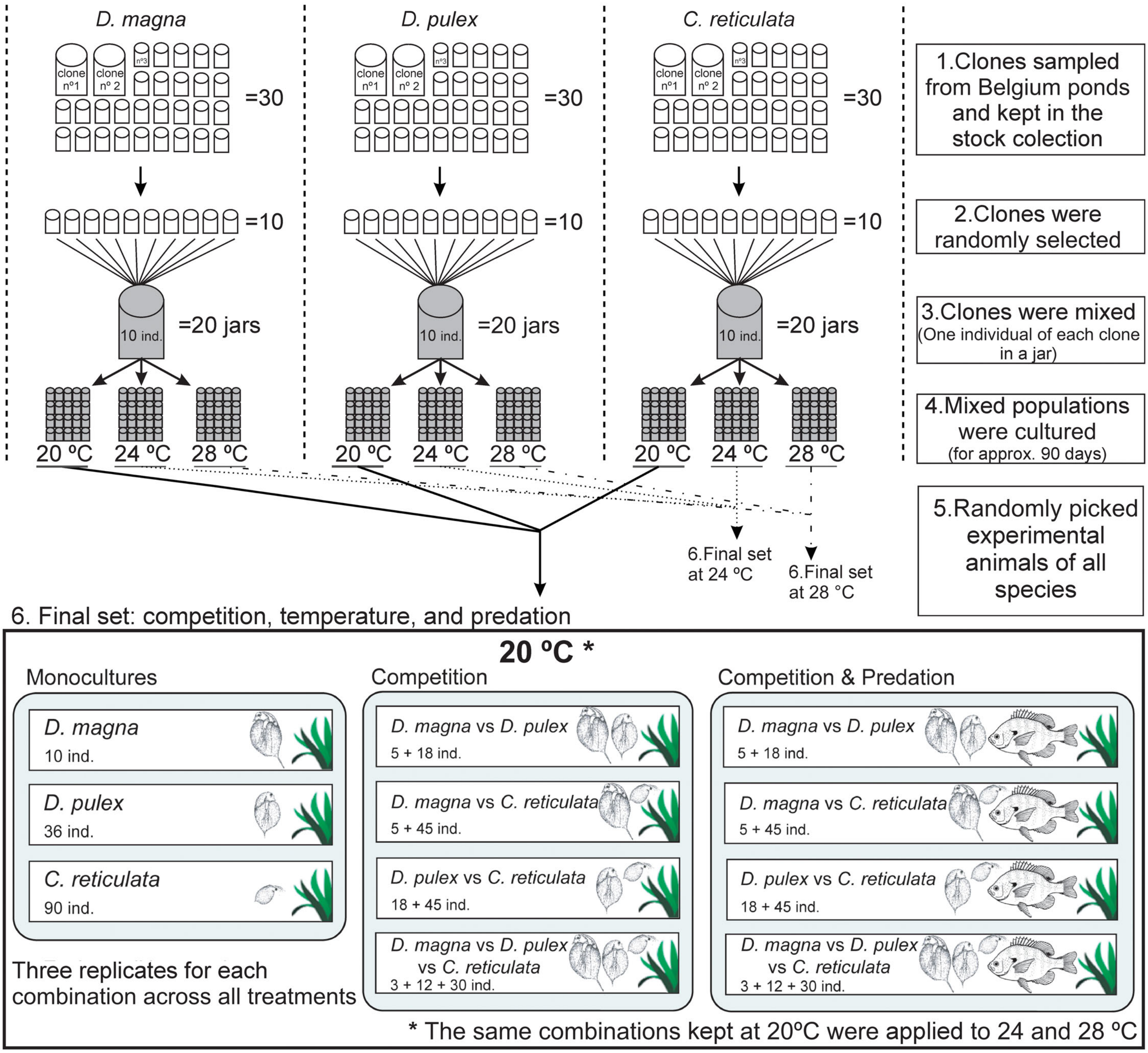

Fig. 1. Schematic overview showing the process from the animals in stock culture to the actual experiment with monocultures as well as two- and three-species communities crossed with temperature (three levels) and predation (presence/absence of fish). The number of individuals (ind.) of each species put in each aquarium at the start of the experiment was adjusted to the differences in body mass so that all units were started with the same total biomass. All treatment combinations were run in triplicate.

Effects of warming and competition on species-specific biomass

To test our first hypothesis on the effect of warming on species-specific biomasses, we applied a two-way type II ANOVA (package "car"; [Fox et al. 2019]) on the data of the three species cultured in monoculture testing for the effects of species, temperature, and their interaction on population biomass. To test our second hypothesis on the effects of warming, competition, and their interaction on each species, we applied three separate two-way type II ANOVAs (one for each species), comparing the biomass of monocultures of a given species to the species-specific biomass of the same species in the different competition trials at three temperatures to assess to what extent the presence of other species influences species-specific biomasses and whether this is temperature dependent.

Effects of warming, predation, and competition on species-specific biomass

To test our third hypothesis that predation has a stronger negative impact on biomass of larger than of smaller species and that this effect is stronger at higher than at low lower temperatures, we performed separate analyses for each species. For two species (*D. pulex* and *C. reticulata*) a three-way ANOVA was carried out to test for the effect of temperature, competition (three levels; no monocultures), and the presence of fish on the species-specific biomass. We square root transformed the biomass data to comply with model assumptions. The data of *D. magna* contained several zeros in the predation treatments; therefore, we ran a GLM (generalized linear model) applying the quasi-Poisson distribution and "log" link function using the "zeroinfl" function from the "pscl" package (Zeileis et al. 2008; Jackman 2020).

To test the hypothesis that predation pressure changes community composition towards the dominance of small-bodied species, we used PERMANOVA (Permutational ANOVA; package "vegan" (Oksanen et al. 2019) testing for differences in community composition among treatment combinations. We carried out three PERMANOVA analyses that excluded data on the monocultures. The first PERMANOVA tested for the effects of factorial combinations of temperature, competition, and predation on all community data. Two additional analyses tested for the effects of temperature and competition on the subsets of data in the absence and presence of fish predation separately. We visualized community composition at the end of the experiment in all competition trials using a ternary plot using species-specific biomass as input data (Hamilton and Ferry 2018).

All analyses were performed in R version 4.0.2 and RStudio version 1.3.1093 (R Core Team 2020); R packages "ggplot2" (Wickham et al. 2019) and "ggtern" (Hamilton and Ferry 2018) were used for graphs.

Results

Effects of warming on species biomass

The population biomass in monocultures differed significantly between species and experimental temperatures ($p = 0.04$ and 0.004, respectively; Table 1). These two factors did not interact, showing that the species responded similarly to warming. The highest biomass values in monoculture when factoring in all species together were observed at 20°C, while 24°C and 28°C did not differ (Fig. 2; post hoc comparisons are shown in Table S3). Irrespective of the thermal regime, the biomass in the *D. magna* monocultures differed significantly from that of *C. reticulata* monocultures; the biomass of *D. pulex* monocultures did not differ from that of the other two species (Fig. 2; Table S3).

Table 1. Output of ANOVA testing whether biomass in monocultures differs among species (*D. magna*, *D. pulex*, and *C. reticulata*), temperature, and their interaction. Df = degrees of freedom. Significant *p*-values (<0.05) are indicated in bold.

	Df	*F*-value	*p*-value
Species	2	3.911	**0.038**
Temperature	2	7.469	**0.004**
S : T	4	1.413	0.269
Residuals	18	–	–

Effects of warming and competition on species biomasses

The biomass of *D. magna* was, in the absence of predation, impacted by the interaction between competition and warming ($p = 0.030$; Table 2, Fig. 3a). Analyzing the isolated effect of warming, post hoc tests showed that *D. magna* reduced biomass from 20°C to both 24 and 28°C ($p = 0.04$ and $p = 0.001$; Table S4). In the post hoc tests for the isolated effect of competition, *D. magna* biomass was significantly reduced from the obtained value in monoculture when competing with *D. pulex* and in the three-species communities ($p = 0.035$ and $p < 0.001$; Fig. 3a and Table S4). When analyzing the interaction between warming and competition, the reduction in *D. magna* in the three-species community compared to in monoculture was significant at 20°C but not at the other two temperatures ($p = 0.004$; Table S4). There is, only at 20°C, also a significant difference between the biomass of *D. magna* when grown together with *C. reticulata* and the biomass of *D. magna* in the three-species communities ($p = 0.003$, Table S4). Another interaction between competition and temperature is revealed by the fact that the biomass of *D. magna* is reduced when grown together with *C. reticulata* at 28 but not at 20°C ($p = 0.006$, Table S4).

D. pulex biomass was negatively influenced by temperature and competition but not by their interaction ($p < 0.001$ for

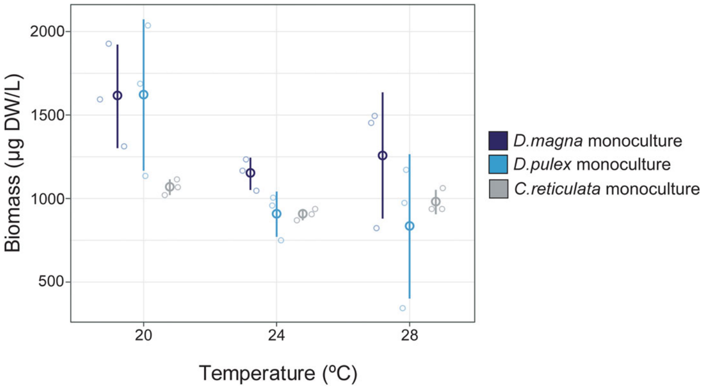

Fig. 2. Mean biomass ± 1 standard deviation in monoculture for *D. magna*, *D. pulex*, and *C. reticulata* at the three experimental temperatures.

Table 2. Output of ANOVA on species biomasses with temperatures (20°C, 24°C, and 28°C) and competition (mono, pairwise 1, pairwise 2, and triple) as dependent variables. Df = degrees of freedom. Significant *p*-values (< 0.05) are indicated in bold.

		Df	F-value	*p*-value
D. magna	Temperature	2	9.813	**<0.001**
	Competition	3	9.114	**<0.001**
	T : C	6	2.862	**0.030**
	Residuals	24	–	–
D. pulex	Temperature	2	15.46	**<0.001**
	Competition	3	13.87	**<0.001**
	T : C	6	0.488	0.810
	Residuals	24		
C. reticulata	Temperature	2	1.974	0.161
	Competition	3	147.8	**<0.001**
	T : C	6	1.042	0.423
	Residuals	24	–	–

both; Table 2). Across all community contexts, *D. pulex* had a lower biomass at both 24 and 28°C than at 20°C ($p = 0.003$ and $p < 0.001$, respectively; Fig. 3a and Table S4). *D. pulex* biomass did not differ between monoculture and in competition with *C. reticulata* at all three temperatures (Fig. 3a and Table S4), but was significantly reduced in competition with *D. magna* and in the three-species communities at all three temperatures (all pairwise comparisons $p < 0.001$; Table S4).

C. reticulata biomass was not affected by temperature but was significantly affected by competition ($p < 0.001$; Table 2). The biomass of this species was significantly higher in monocultures than in all multi-species communities (all pairwise comparisons $p < 0.001$; Table S4).

Effect of temperature, competition, and predation on species-specific biomass

The effect of 10 min of fish predation on a weekly interval was strong and highly significant (Fig. 3b and Table 3). Fish predation had a strong negative effect on *D. magna* biomass and reduced all populations of this species to very low numbers, removing it from most aquaria ($p < 0.001$, Table 3 and Fig. 4). There was also a significant interaction effect between temperature and competition, independent of predation treatment (all $p < 0.001$, Table 3): *D. magna* had significantly reduced biomass at high temperatures (24°C and 28°C) in combination with *D. pulex* and in the three species combination compared to 20°C in combination with *C. reticulata* ($p < 0.001$, Table 3 and Fig. 3b).

In general, the *D. pulex* populations were also strongly influenced by temperature, competition, fish predation (all $p < 0.001$; Fig. 4 and Table 3), and by the interaction of predation and competition ($p = 0.010$; Fig. 3b and Table 3). Fish predation decreased *D. pulex* biomass in all competition trials (all $p < 0.002$; Table S5). In the presence of predation, *D. pulex* biomass did not differ between competition trials, whereas in the absence of fish, the biomass of *D. pulex* in combination with *D. magna* and in the three-species community was significantly lower than in the competition trial with *C. reticulata* (both $p < 0.001$; Table S5; Fig. 4).

Generally, the biomass of *C. reticulata* was higher in the presence of fish ($p = 0.012$; Table 3 and Fig. 4), and higher when placed together with *D. magna* in comparison when under competition with *D. pulex* ($p = 0.04$) and in the three-species community ($p < 0.001$; Table 3 and Fig. 4). Results of post hoc tests are shown in the Table S5.

Effect of temperature, competition, and predation on community composition

The community composition in the aquaria with two- and three-species communities was significantly altered by fish predation ($p = 0.001$), temperature ($p = 0.005$), and competition ($p = 0.001$, Table 4). Predation also interacted with competition and temperature to determine community composition (Table 4). In absence of predation, community composition was significantly affected by competition, temperature, and their interaction ($p = 0.002$, Table 4). In the presence of fish, only competition significantly affected community composition ($p = 0.001$). The ternary plot shows that fish predation led to the exclusion of *D. magna* (i.e., 0 values for *D. magna* in the DmxCr, DmxDp, and DmxDpxCr treatments). Predation shifted the three-species community more toward the dominance of *C. reticulata* in the 24°C and 28°C temperature treatments, and toward the dominance of *D. pulex* in the 20°C treatment (Fig. 5). In the absence of predation, *C. reticulata* was largely excluded in all competition trials. In treatments where *D. magna* and *D. pulex* coexisted (i.e., DmxDp and DmxDpxCr), *D. magna* dominance was stronger at higher than at lower temperatures.

Discussion

As expected, warming negatively impacted the biomass of all three studied zooplankton species in monoculture, and this translated also into lower biomasses of all species in the warming treatments in two- or three-species communities. The large- and medium-sized *Daphnia* species were more negatively affected by warming than the smaller *Ceriodaphnia* species. However, diverging from our expectations, warming alone did not alter the relative dominance of large- vs. small-bodied cladocerans in competition trials. Predation had a strong negative impact on the biomass of larger-bodied *Daphnia* but not on the biomass of the smaller-bodied *Ceriodaphnia*. In line with our expectations, predation by fish did favor the relative biomass of small-bodied *Ceriodaphnia* in the competition trials, and this effect was enhanced at higher temperatures. The observed responses to warming, competition, and predation patterns are suggestive of a strong body size-mediated effect of these drivers, but with some important

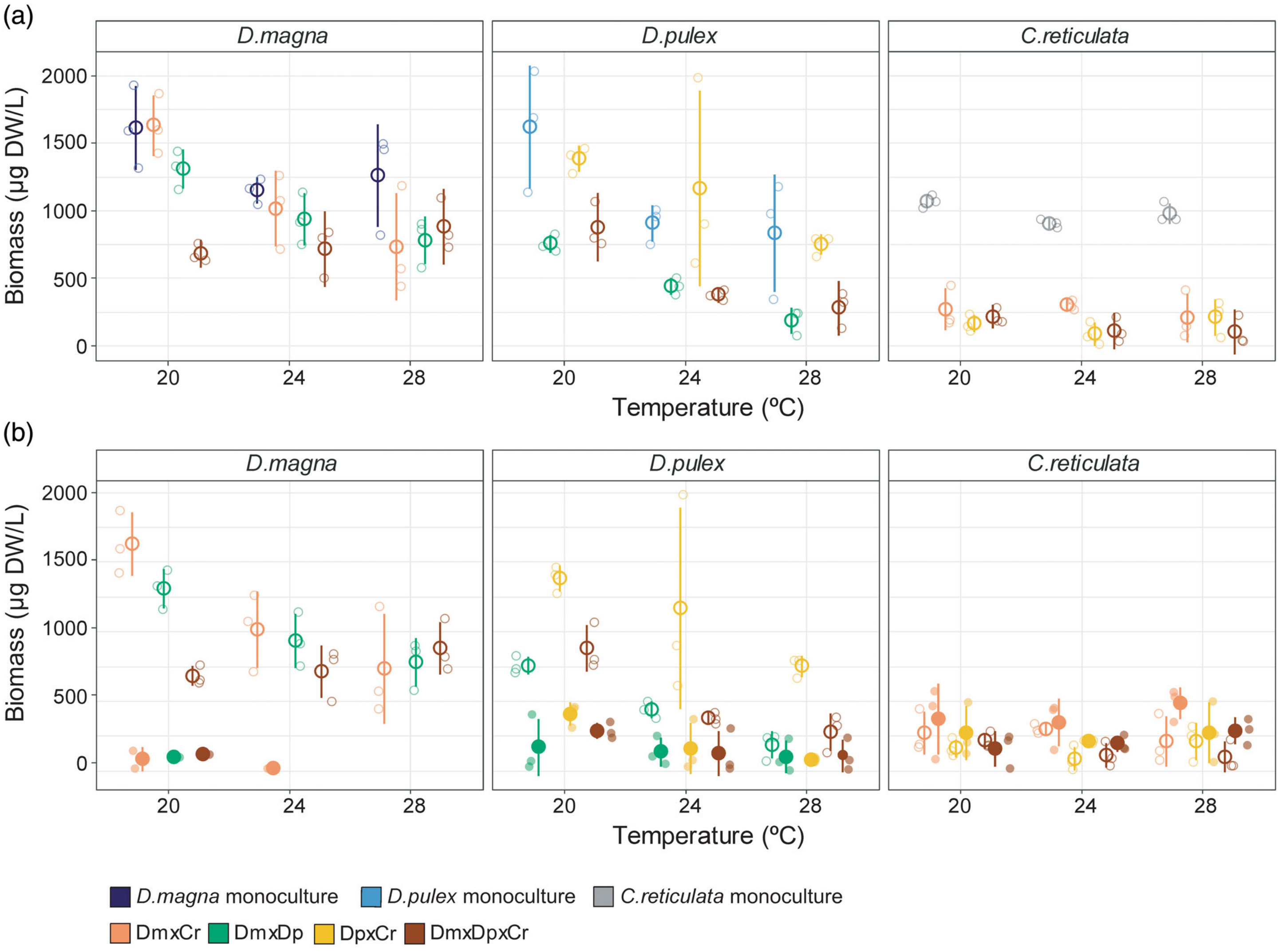

Fig. 3. Mean species-specific biomass ± 1 standard deviation for *D. magna*, *D. pulex*, and *C. reticulata* at the three experimental temperatures. (**a**) Biomass in the absence of predation, comparing values from monoculture, two pairwise competition trials, and the three-species community. (**b**) Comparison of the biomass under competition in the absence (empty symbols) and in the presence of predation (filled symbols). Dm: *D. magna*, Dp: *D. pulex*, and Cr: *C. reticulata*.

deviations that are linked to the higher heat tolerance of the large-bodied *D. magna* compared to the medium-sized *D. pulex*. In the following paragraphs, we discuss these results in more detail and put them in context.

Effect of warming

Sustaining higher metabolic demands at higher temperatures both in terms of food and oxygen has proven to be more challenging in larger compared to smaller animals, especially in aquatic ectotherms (Goulden et al. 1982; Atkinson 1994). Increased temperatures are thus expected to cause higher stress levels and metabolic losses in larger species than in smaller ones and to reduce individual biomass production (Kooijman 2010). This latter prediction was only partially confirmed by our results. We did observe that the population biomass of the smallest of the three species tested, *C. reticulata*, was less impacted by warming than that of the larger-bodied *Daphnia* species. Yet, among the two *Daphnia* species, *D. pulex* suffered stronger reductions in biomass when cultured at 28°C instead of 20°C than *D. magna*. While studies on the effects of body-size variation within *Daphnia* species showed that smaller animals have a higher heat tolerance (Geerts et al. 2015; Brans et al. 2017), thermal tolerance is not only dependent on body size, but also depends on physiological characteristics such as general stress defenses (e.g., heat-shock proteins) and hemoglobin content (Gerke et al. 2011; Zeis 2020). Indeed, using a different set of clones, Vanvelk et al. (2021) observed that *D. magna* has a higher heat tolerance (CTmax and growth) than other *Daphnia* species (*D. pulicaria* and *D. galeata*), which is in line with our observation that *D. magna* performed better at high temperatures than *D. pulex*. Several studies showed that *D. magna* can increase its heat tolerance both via phenotypic plasticity and adaptive evolutionary changes in morphology and stress physiology (Van Doorslaer et al. 2009; Geerts et al. 2015; Brans et al. 2017; Vanvelk et al. 2021).

Warming and competition

Large-bodied zooplankton species generally have a competitive dominance over smaller ones. This is because large-bodied species can feed on broader particle size ranges (DeMott 1989), are more energy-efficient (Gliwicz 1980), more starvation resistant (Tessier et al. 1983), and can sometimes directly compete with smaller-bodied species through interference competition (Gliwicz 1980; Goulden et al. 1982; Ye et al. 2013). Our results are largely in line with an expected asymmetric competitive effect among differently-sized Daphniidae species. *Ceriodaphnia* is strongly suppressed by both *Daphnia* species in any combination of two- or three-species communities. The two *Daphnia* species do suppress each other's population biomasses, but *D. magna* has a stronger impact on *D. pulex* than *D. pulex* on *D. magna*. We observed, however, an interaction with temperature that is linked to the different sensitivity to warming of the three species. The resulting pattern is that in the absence of fish, *D. magna* dominates in all community contexts in which it was inoculated and that the dominance of *D. magna* in competition with *D. pulex* is slightly stronger at higher compared to lower temperatures (Fig. 3a). This reflects again that *D. pulex* is more negatively affected by warming than *D. magna* despite being smaller-bodied.

Warming and predation

There is a vast amount of evidence for the pervasive influence of fish predation on zooplankton densities and body size distribution (Brooks and Dodson 1965; Lampert and Sommer 2011). Fish are visual predators and can impact the size distribution of zooplankton by preferentially removing large-bodied prey species that are visually more conspicuous, shifting the age distribution to younger animals, inducing a reduction in body size through phenotypic plasticity, and driving evolution by selection against larger-bodied genotypes (Iglesias et al. 2011; Šorf et al. 2015; Santangelo et al. 2018). In our experiment, predation resulted in a profound shift in species composition of the cladoceran communities towards the dominance of the small-bodied species. The large-bodied *D. magna* was driven to near-extinction, while the small-bodied *Ceriodaphnia* was positively affected in terms of biomass by the fish presence. The latter was likely due to competitive release from *D. magna*.

Table 3. Output of GLM for biomasses of *D. magna* and ANOVAs for biomasses of *D. pulex* and *C. reticulata* testing for the effect of temperature (20°C, 24°C, and 28°C), competition (all pairwise and the three-species communities), and predation (fish or no fish) Df = degrees of freedom. Significant *p*-values (< 0.05) are indicated in bold. The intercept in the GLM model for pairwise comparison is 20°C, DmxCr, and the presence of fish predation.

		Estimate	*z*-value	*p*-value
D. magna	Intercept	7.39	520.04	<**0.001**
	Temperature (24°C)	−0.48	−20.99	<**0.001**
	Temperature (28°C)	−0.79	−30.93	<**0.001**
	Competition (DmxDp)	−0.21	−9.84	<**0.001**
	Competition (DmxDpxCr)	−0.83	−32.23	<**0.001**
	Predation (no fish)	−2.75	−54.17	<**0.001**
	T (24°C) : C (DmxDp)	0.14	4.24	<**0.001**
	T (28°C) : C (DmxDp)	0.27	7.51	<**0.001**
	T (24°C) : C (DmxDpxCr)	0.49	12.89	<**0.001**
	T (28°C) : C (DmxDpxCr)	1.01	26.25	<**0.001**

		Df	*F*-value	*p*-value
D. pulex	Temperature	2	18.93	<**0.001**
	Competition	2	12.41	<**0.001**
	Predation	1	98.64	<**0.001**
	T : C	4	0.492	0.742
	T : P	2	1.369	0.267
	C : P	2	5.219	**0.010**
	T : C : P	4	0.806	0.529
	Residuals	36	–	–
C. reticulata	Temperature	2	0.442	0.646
	Competition	2	5.635	**0.007**
	Predation	1	7.038	**0.012**
	T : C	4	0.250	0.908
	T : P	2	1.342	0.274
	C : P	2	0.060	0.942
	T : C : P	4	1.070	0.383
	Residuals	36	–	–

Our results are relevant with respect to the global distribution of Daphniidae species. It is well-documented that large *Daphnia* species are rare in lowland tropics where the water bodies are strongly dominated by small zooplankton species (Sarma et al. 2005; Havens et al. 2014). In subtropical, temperate, and arctic regions, however, *Daphnia* are widespread (Smirnov et al. 1995; Sarma et al. 2005; Meerhoff et al. 2007). This has been attributed to the fact that large-bodied ectothermic species are at a disadvantage at higher

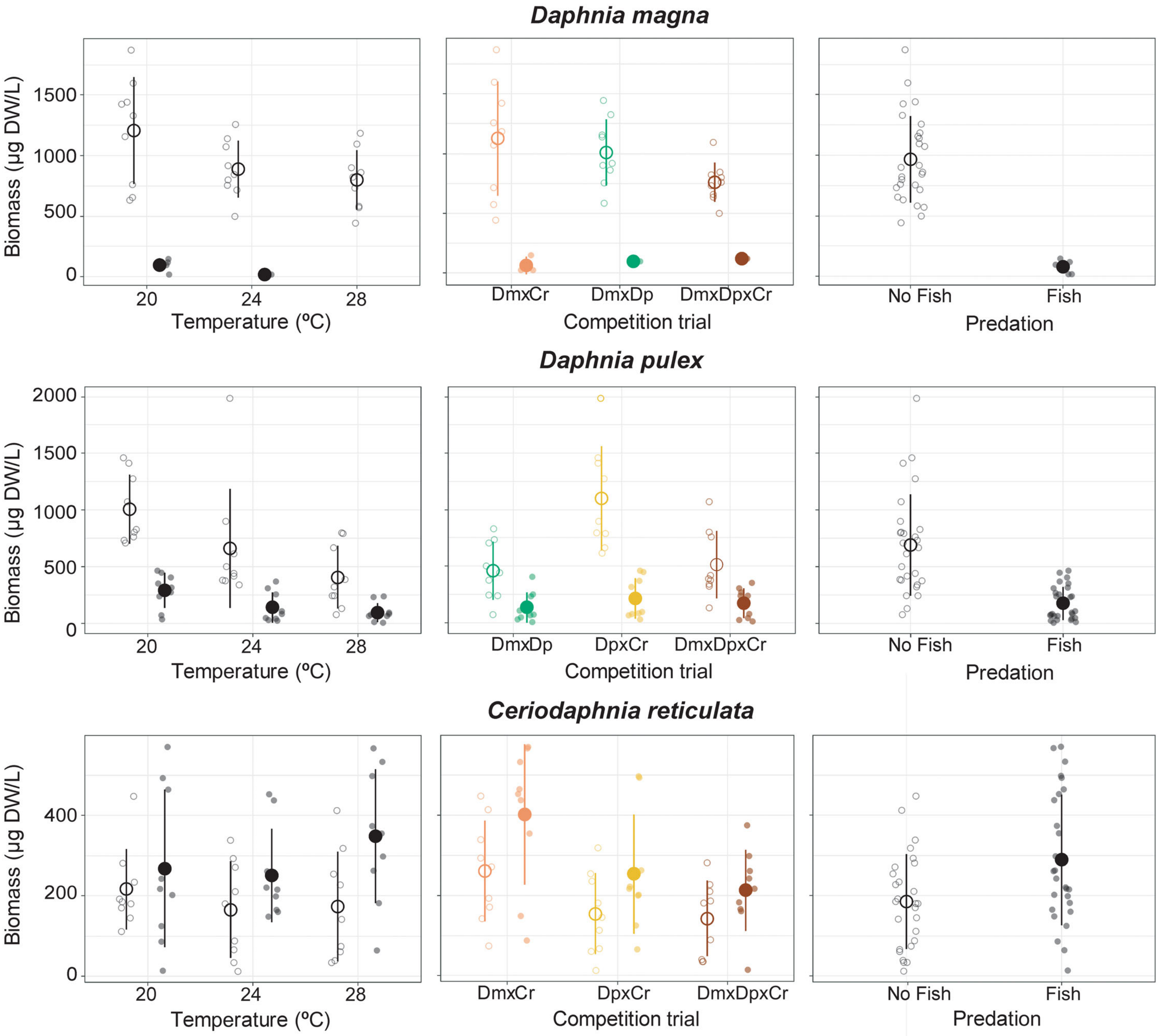

Fig. 4. Main effects for model testing differences in species-specific biomass across temperature (left), competition trial (middle), and predation treatment (right). The mean species-specific biomasses ± 1 standard deviation are shown for *D. magna* (upper), *D. pulex* (middle), and *C. reticulata* (lower). Empty symbols represent the absence of predation and filled symbols the presence of predation.

temperatures because of their higher metabolic demands (Lear et al. 2020) as well as to the sustained high levels of size-selective predation by fish in tropical lowland waters (Iglesias et al. 2017; He et al. 2018). Our results confirm that both have an impact, but that fish predation is likely the key factor shifting cladoceran body size distribution to the dominance of very small individuals (Meerhoff et al. 2007; Iglesias et al. 2017).

Implications

Predicting responses to global change requires mechanistic insights in physiological responses and species interactions, among others (Urban et al. 2016). In the present study, we documented the link between warming and two ecological interactions in simplified cladoceran communities. Warming and competition did not move community composition away from a strong dominance by large-bodied *Daphnia*.

Table 4. Results of the permutational ANOVA (PERMANOVA) on the community composition at the end of the microcosm experiment testing for the effect of temperature and competition. Df = degrees of freedom and SS = the sum of the squares. Significant *p*-values (< 0.05) are indicated in bold, and marginally significant values (< 0.1) are underlined.

	Df	SS	R^2	*F*-value	*p*-value
PERMANOVA 1					
Temperature	2	0.435	0.024	3.092	**0.005**
Competition	3	5.903	0.326	27.979	**0.001**
Predation	1	5.418	0.299	77.04	**0.001**
T : C	6	0.305	0.0168	0.723	0.790
T : P	2	0.423	0.023	3.004	**0.007**
C : P	3	1.719	0.095	8.151	**0.001**
T : C : P	6	0.539	0.023	1.279	0.209
Residuals	48	3.376	0.186	–	–
Total	71	18.119	1.000	–	–
PERMANOVA 2: no fish					
Temperature	2	0.360	0.024	8.863	**0.001**
Competition	6	13.484	0.885	110.641	**0.001**
T : C	12	0.544	0.036	2.231	**0.002**
Residuals	42	0.853	0.056	–	–
Total	62	15.24	1.000	–	–
PERMANOVA 3: Fish					
Temperature	2	0.517	0.067	2.187	0.052
Competition	3	3.832	0.500	10.814	**0.001**
T : C	6	0.514	0.067	0.725	0.763
Residuals	24	2.835	0.368	–	–
Total	35	7.698	1.000	–	–

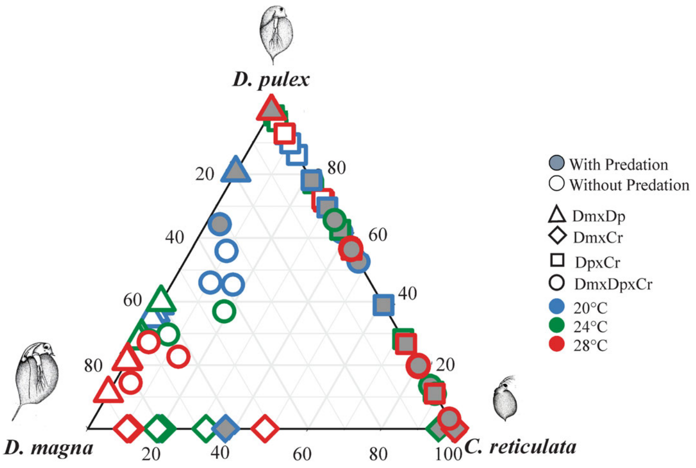

Fig. 5. Ternary plot visualizing the resulting community composition (% of biomasses) under the different treatments of temperature, predation, and competition trials. Dm: *D. magna*; Dp: *D. pulex*; Cr: *C. reticulata*.

Only under fish predation did community composition shift completely to the dominance of the small-bodied *Ceriodaphnia*, and this effect was enhanced by warming. Most of our observations are in line with expectations derived from differences in the average body size of the studied species. Others deviated from these predictions, such as the stronger suppression of *D. pulex* than *D. magna* biomass at higher temperatures.

The strong shift to dominance of small-bodied zooplankton upon warming in the presence of fish can have important implications for ecosystem structure and functioning. It may induce changes in food web structure and lead to less efficient energy transfer and nutrient cycling (Barnett et al. 2007; O'Connor et al. 2009). Large-bodied *Daphnia* species such as *D. magna* and *D. pulex* are keystone species that are very efficient in grazing on unicellular algae, and thus can contribute to the prevention of (toxic) algae blooms (Lampert and Sommer 2011; Chislock et al. 2013; Gianuca et al. 2016). Their replacement by smaller-bodied taxa will likely reduce top-down control of algae and may contribute to a more frequent occurrence of algal blooms, especially since warming also stimulates algal growth and dominance by cyanobacteria (Kosten et al. 2012), ultimately leading to turbid, algae-dominated systems. This also points to an observation that in our study, we quantified the interactive effects of warming, competition, and predation in a setting characterized by good-quality food. Given that warming is expected to enhance the relative and absolute abundance of cyanobacteria and other inedible or less-quality food (Kosten et al. 2012; Visser et al. 2016), it will be important in future work to also explore how low-quality food would impact the observed community shifts upon warming, competition, and predation. Colony-forming and toxic cyanobacteria can increase the mortality of large-bodied cladocerans (Moustaka-Gouni and Sommer 2020), which could potentially reduce the dominance of large-bodied *Daphnia* in the zooplankton a community under higher temperatures even in the absence of fish predation.

Data availability statement

The data that support the findings of this study are available from the corresponding author, F.F.B., upon reasonable request.

References

Adamczuk, M. 2010. Different life-history trade-offs of two daphnia species (Cladocera, crustacea) under natural conditions as the response to predation and competition. Ann. Limnol. Int. J. Limnol. **46**: 241–247. doi:10.1051/limn/2010022

Atkinson, D. 1994. Temperature and organism size-a law for ectotherms? Adv Ecol **25**: 1–58.

Atkinson, D., S. A. Morley, D. Weetman, and R. N. Hughes. 2001. Offspring size responses to maternal temperature in ectotherms. Anim Dev Ecol **14**: 269–285.

Barnett, A. J., K. Finlay, and B. E. Beisner. 2007. Functional diversity of crustacean zooplankton communities: Towards a trait-based classification. Freshw Biol **52**: 796–813. doi:10.1111/j.1365-2427.2007.01733.x

Bomfim, F. d. F., M. G. G. Melão, R. C. Gebara, and F. A. Lansac-Tôha. 2022. Warming alters the metabolic rates and life-history parameters of *Ceriodaphnia silvestrii* (Cladocera). An Brazil Acad Sci **94**: e20200604.

Brans, K. I., M. Jansen, J. Vanoverbeke, N. Tüzün, R. Stoks, and L. de Meester. 2017. The heat is on: Genetic adaptation to urbanization mediated by thermal tolerance and body size. Glob. Chang. Biol. **23**: 5218–5227. doi:10.1111/gcb.13784

Brans, K. I., J. M. T. Engelen, C. Souffreau, and L. de Meester. 2018. Urban hot-tubs: Local urbanization has profound effects on average and extreme temperatures in ponds. Landsc Urban Plan **176**: 22–29. doi:10.1016/j.landurbplan.2018.03.013

Brooks, J. L., and S. I. Dodson. 1965. Predation, body size and composition of plankton. Science **1979**: 28–35.

Chase, J. M., and others. 2002. The interaction between predation and competition: A review and synthesis. Ecol. Lett. **5**: 302–315. doi:10.1046/j.1461-0248.2002.00315.x

Chislock, M. F., O. Sarnelle, L. M. Jernigan, and A. E. Wilson. 2013. Do high concentrations of microcystin prevent daphnia control of phytoplankton. Water Res. **47**: 1961–1970.

Daufresne, M., K. Lengfellner, and U. Sommer. 2009. Global warming benefits the small in aquatic ecosystems. Proc. Natl. Acad. Sci. **106**: 12788–12793.

De Meester, L., R. Stoks, and K. I. Brans. 2018. Genetic adaptation as a biological buffer against climate change : Potential and limitations. Integr Zool **13**: 372–391. doi:10.1111/1749-4877.12298

DeMott, W. R. 1989. The role of competition in zooplankton succession, p. 378. *In* U. Sommer [ed.], Plankton ecology: Succession in planktonic communities. Springer-Verlag.

Ersoy, Z., and others. 2017. Size-based interactions and trophic transfer efficiency are modified by fish predation and cyanobacteria blooms in Lake Mývatn, Iceland. Freshw Biol **62**: 1942–1952. doi:10.1111/fwb.13039

Forster, J., A. G. Hirst, and G. Woodward. 2011. Growth and development rates have different thermal responses. Am Nat. **178**: 668–678.

Forster, J., A. G. Hirst, and D. Atkinson. 2012. Warming-induced reductions in body size are greater in aquatic than terrestrial species. Proc. Natl. Acad. Sci. U. S. A. **109**: 19310–19314. doi:10.1073/pnas.1210460109

Fox, J., and others. 2019. Car-Companion to Applied Regression, R program. Sage, p. 149.

Geerts, A. N., and others. 2015. Rapid evolution of thermal tolerance in the water flea daphnia. Nat Clim Chang **5**: 665–668. doi:10.1038/nclimate2628

Gerke, P., C. Börding, B. Zeis, and R. J. Paul. 2011. Adaptive haemoglobin gene control in *Daphnia pulex* at different oxygen and temperature conditions. Comp Biochem Physiol A Mol Integr Physiol **159**: 56–65. doi:10.1016/j.cbpa.2011.01.017

Gianuca, A. T., J. H. Pantel, and L. De Meester. 2016. Disentangling the effect of body size and phylogenetic distances on zooplankton top-down control of algae. Proc R Soc B Biol Sci **283**: 20160487. doi:10.1098/rspb.2016.0487

Glazier, D. S., and P. Calow. 1992. Energy allocation rules in *Daphnia magna*: Clonal and age differences in the effects of food limitation. Oecologia **90**: 540–549.

Gliwicz, Z. M., 1980. Filtering rates, food size selection, and feeding rates in cladocerans – another aspect of interspecific competition, p. 282–291. *In* W. C. Kerfoot [ed.], Evolution and ecology of zooplankton communities. Univ. Press of New England.

Goulden, C. E., L. L. Henry, and A. J. Tessier. 1982. Body size, energy reserves, and competitive ability in three species of Cladocera. Ecology **63**: 1780–1789. doi:10.2307/1940120

Hamilton, N. E., and M. Ferry. 2018. Ggtern: Ternary diagrams using ggplot2. J Stat Softw **87**: 1–17. doi:10.18637/jss

Hart, R. C., and E. A. Bychek. 2011. Body size in freshwater planktonic crustaceans: An overview of extrinsic determinants and modifying influences of biotic interactions. Hydrobiologia **668**: 61–108. doi:10.1007/s10750-010-0400-y

Havens, K. E., and others. 2014. Temperature effects on body size of freshwater crustacean zooplankton from Greenland to the tropics. Hydrobiologia **743**: 27–35. doi:10.1007/s10750-014-2000-8

He, H., H. Jin, E. Jeppesen, K. Li, Z. Liu, and Y. Zhang. 2018. Fish-mediated plankton responses to increased temperature in subtropical aquatic mesocosm ecosystems: Implications for lake management. Water Res. **144**: 304–311. doi:10.1016/j.watres.2018.07.055

Hu, S. S., and A. J. Tessier. 1995. Seasonal succession and the strength of intra- and interspecific competition in a daphnia assemblage. Ecology **76**: 2278–2294.

Iglesias, C., and others. 2011. High predation is of key importance for dominance of small-bodied zooplankton in warm shallow lakes: Evidence from lakes, fish exclosures and surface sediments. Hydrobiologia **667**: 133–147. doi:10.1007/s10750-011-0645-0

Iglesias, C., and others. 2017. Fish but not macroinvertebrates promote trophic cascading effects in high density submersed plant experimental lake food webs in two contrasting climate regions. Water (Switzerland) **9**: 514.

IPCC. 2014. Climate change 2014: Synthesis report. Contribution of working groups I, II and III to the fifth assessment report of the intergovernmental panel on climate change, p. 1–112. *In* C. W. Team, R. K. Pachauri, and L. A. Meyer [eds.], v. **151**. IPCC. doi:10.1017/CBO9781107415324

Jackman, S. 2020. Pscl: Classes and methods for R developed in the political science computational laboratory. United States Studies Centre, University of Sydney.

Jeppesen, E., and others. 2014. Climate change impacts on lakes: An integrated ecological perspective based on a multi-faceted approach, with special focus on shallow lakes. J. Limnol. **73**: 88–111. doi:10.4081/jlimnol.2014.844

Jiang, X., W. Yang, L. Zhang, L. Chen, and Y. Niu. 2014. Predation and cyanobacteria jointly facilitate competitive dominance of small-bodied cladocerans. J. Plankton Res. **36**: 956–965. doi:10.1093/plankt/fbu032

Kooijman, S. A. L. M. 2010. Summary of concepts of dynamic energy budget theory. Cambridge University Press, p. 1–72.

Kosten, S., and others. 2012. Warmer climates boost cyanobacterial dominance in shallow lakes. Glob. Chang. Biol. **18**: 118–126.

Lampert, W., and U. Sommer. 2011. Limnoecology. Oxford Univ. Press.

Lear, K. O., D. L. Morgan, J. M. Whitty, N. M. Whitney, E. E. Byrnes, S. J. Beatty, and A. C. Gleiss. 2020. Divergent field metabolic rates highlight the challenges of increasing temperatures and energy limitation in aquatic ectotherms. Oecologia **193**: 311–323. doi:10.1007/s00442-020-04669-x

Lenth, R., H. Singmann, J. Love, P. Buerkner, and M. R. L. M. Herve. 2019. Emmeans: Estimated marginal means, aka least-squares means. Am Stat **34**.

Meerhoff, M., J. M. Clemente, F. T. de Mello, C. Iglesias, A. R. Pedersen, and E. Jeppesen. 2007. Can warm climate-related structure of littoral predator assemblies weaken the clear water state in shallow lakes? Glob. Chang. Biol. **13**: 1888–1897. doi:10.1111/j.1365-2486.2007.01408.x

Moore, M. V., C. L. Folt, and R. S. Stemberger. 1996. Consequences of elevated temperatures for zooplankton assemblages in temperate lakes. Arch Hydrobiol **135**: 289–319. doi:10.1127/archiv-hydrobiol/135/1996/289

Moustaka-Gouni, M., and U. Sommer. 2020. Effects of harmful blooms of large-sized and colonial cyanobacteria on aquatic food webs. Water (Switzerland) **12**: 1–19. doi:10.3390/W12061587

Norberg, J., M. C. Urban, M. Vellend, C. A. Klausmeier, and N. Loeuille. 2012. Eco-evolutionary responses of biodiversity to climate change. Nat Clim Chang **2**: 747–751. doi:10.1038/nclimate1588

O'Connor, M. I., M. F. Piehler, D. M. Leech, A. Anton, and J. F. Bruno. 2009. Warming and resource availability shift food web structure and metabolism. PLoS Biol. **7**: 3–8. doi:10.1371/journal.pbio.1000178

Oksanen, J., and others. 2019. Vegan Community Ecology Package. R Package Version **296**.

Parmesan, C. 2006. Ecological and evolutionary responses to recent climate change. Annu. Rev. Ecol. Evol. Syst. **37**: 637–669. doi:10.1146/annurev.ecolsys.37.091305.110100

Power, M. E., and J. H. Todd. 1976. Effects of increasing temperature on social behaviour in territorial groups of pumpkinseed sunfish, *Lepomis Gibbosus*. Environ. Pollut. **10**: 217–223.

R Core Team. 2020. A language and environment for statistical computing. R Core Team, https://www.r-project.org

Rosset, V., A. Lehmann, and B. Oertli. 2010. Warmer and richer? Predicting the impact of climate warming on species richness in small temperate waterbodies. Glob. Chang. Biol. **16**: 2376–2387.

Santangelo, J. M., B. N. Soares, T. Paes, P. Maia-Barbosa, R. Tollrian, and R. L. Bozelli. 2018. Effects of vertebrate and invertebrate predators on the life history of *Daphnia similis* and *Moina macrocopa* (crustacea: Cladocera). Ann. Limnol. Int. J. Limnol. **54**: 25. doi:10.1051/limn/2018015

Sarma, S. S. S., S. Nandini, and R. D. Gulati. 2005. Life history strategies of cladocerans: Comparisons of tropical and temperate taxa. Hydrobiologia **542**: 315–333. doi:10.1007/s10750-004-3247-2

Scheffers, B. R., and others. 2016. The broad footprint of climate change from genes to biomes to people. Science **1979**: 719–732. doi:10.1126/science.aaf7671

Smirnov, N. N., S. M. Glagolev, N. M. Korovchinsky, A. A. Kotov, M. Ya, Orlova-Bienkowskaja, and I. K. Rivier. 1995. Cladocera (Crustacea), p. 34–74. *In* V. R. Alekseev [ed.], Guides to the identification of the freshwater invertebrates of Russia and surrounding territories. Zoological Institute of Russian Academy of Sciences.

Šorf, M., and others. 2015. Zooplankton response to climate warming: A mesocosm experiment at contrasting temperatures and nutrient levels. Hydrobiologia **742**: 185–203. doi:10.1007/s10750-014-1985-3

Tessier, A. J., L. L. Henry, C. E. Goulden, and M. W. Durand. 1983. Starvation in daphnia: Energy reserves and reproductive allocation. Limnol. Oceanogr. **28**: 667–676. doi:10.4319/lo.1983.28.4.0667

Thakur, M. P., J. N. Griffin, T. Künne, S. Dunker, A. Fanesi, and N. Eisenhauer. 2018. Temperature effects on prey and basal resources exceed that of predators in an experimental community. Ecol. Evol. **8**: 12670–12680. doi:10.1002/ece3.4695

Thébault, E., and M. Loreau. 2003. Food-web constraints on biodiversity–ecosystem functioning relationships. PNAS **100**: 14949–14954.

Urban, M. C., and others. 2016. Improving the forecast for biodiversity under climate change. Science **1979**: 1113–1124. doi:10.1126/science.aad8466

Van Doorslaer, W., R. Stoks, C. Duvivier, A. Bednarska, and L. De Meester. 2009. Population dynamics determine genetic adaptation to temperature in daphnia. Evolution (N Y) **63**: 1867–1878. doi:10.1111/j.1558-5646.2009.00679.x

Vanvelk, H., L. Govaert, E. M. van den Berg, K. L. Brans, and L. De-Meester. 2021. Interspecific differences, plastic and evolutionary responses to a heat wave in three co-occurring daphnia species. Limnol. Oceanogr. **66**: 1201–1220. doi:10.1002/lno.11675

Verberk, W. C. E. P., D. Atkinson, K. N. Hoefnagel, A. G. Hirst, C. R. Horne, and H. Siepel. 2021. Shrinking body sizes in response to warming: Explanations for the temperature–size rule with special emphasis on the role of oxygen. Biol. Rev. **96**: 247–268. doi:10.1111/brv.12653

Visser, P. M., J. M. H. Verspagen, G. Sandrini, L. J. Stal, H. C. P. Matthijs, T. W. Davis, H. W. Paerl, and J. Huisman. 2016. How rising CO2 and global warming may stimulate harmful cyanobacterial blooms. Harmful Algae **54**: 145–159. doi:10.1016/j.hal.2015.12.006

Wickham, H., and others. 2019. Create Elegant Data Visualisations Using the Grammar of Graphics (ggplot2). Springer, p. 227.

Ye, L., C. Y. Chang, C. García-Comas, G. C. Gong, and C. hao Hsieh. 2013. Increasing zooplankton size diversity enhances the strength of top-down control on phytoplankton through diet niche partitioning. J Animal Ecol **82**: 1052–1061. doi:10.1111/1365-2656.12067

Zeis, B. 2020. Hemoglobin in arthropods—Daphnia as a model. *In* U. Hoeger and J. Harris [eds.], Vertebrate and invertebrate respiratory proteins, lipoproteins and other body fluid proteins. Subcellular biochemistry. Springer.

Zeis, B., D. Becker, P. Gerke, M. Koch, and R. J. Paul. 2013. Hypoxia-inducible haemoglobins of *Daphnia pulex* and their role in the response to acute and chronic temperature increase. Biochim Biophys Acta Proteins Proteom **1834**: 1704–1710. doi:10.1016/j.bbapap.2013.01.036

Zeileis, A., C. Kleiber, and S. Jackman. 2008. Regression models for count data in R. J. Stat. Softw. **27**: 1–25.

Zuur, A. F., E. N. Ieno, and C. S. Elphick. 2010. A protocol for data exploration to avoid common statistical problems. Methods Ecol Evol **1**: 3–14. doi:10.1111/j.2041-210x.2009.00001.x

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Conflict of interest

All authors declare no conflicts of interest.

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