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Interactive effects of nutrients and temperature on herbivorous predation in a coastal plankton community

Gayantonia Franzè [1*,a], **Stephanie I. Anderson** [1,b], **Joshua D. Kling** [2], **Paul Wilburn** [3], **David A. Hutchins** [2], **Elena Litchman** [3,c], **Tatiana A. Rynearson** [1], **Susanne Menden-Deuer** [1]

[1]Graduate School of Oceanography, University of Rhode Island, Narragansett, Rhode Island
[2]Department of Biological Sciences, University of Southern California, Los Angeles, California
[3]Michigan State University, Kellogg Biological Station, Hickory Corners, Michigan

Abstract

Marine microbial communities in coastal environments are subject to both seasonal fluctuations and anthropogenic alterations of environmental conditions. The separate influences of temperature and resource-dependency on phytoplankton growth, community, and ecosystem metabolism are relatively well understood. However, winners and losers in the ocean are determined based on the interplay among often rapidly changing biological, chemical and physical drivers. The direct, indirect, and interactive effects of these conditions on planktonic food web structure and function are poorly constrained. Here, we investigated how simultaneous manipulation of temperature and nutrient availability affects trophic transfer from phytoplankton to herbivorous protists, and their resulting implications at the ecosystem level. Temperature directly affected herbivorous protist composition; ciliates dominated (66%) in colder treatment and dinoflagellates (60%) at warmer temperatures. Throughout the experiments, grazing rates were < 0.1 d^{-1}, with higher rates at subzero temperatures. Overall, the nutrient–temperature interplay affected trophic transfer rates antagonistically when nutrients were amended, and synergistically, when nutrients were not added. This interaction resulted in higher percentages of primary production consumed under nutrient unamended compared to nutrient amended conditions. At the ecosystem level, these changes may determine the fate of primary production, with most of the production likely exported out of the pelagic zone in high-temperature and nutrient conditions, while high-temperature and low-nutrient availability strengthened food web coupling and enhanced trophic transfer. These results imply that in warming oceans, management of coastal nutrient loading will be a critical determinant of the degree of primary production removal by microzooplankton and dependent ecosystem production.

Plankton play a fundamental role in marine ecosystems, controlling major elemental cycles and supporting nearly all marine food webs. Their response to rapidly shifting environmental conditions can reshape microbial networks impacting elemental and biological cycles and the magnitude of primary production (Hutchins and Fu 2017). This particularly applies to coastal waters in temperate regions which are subject to both large-scale seasonal changes and increasingly, fluctuating stressors that can alter marine communities and, consequently, affect ecosystem services essential to humans (Marrec et al. 2021).

*Correspondence: franze@hi.no

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

[a]Present address: Institute of Marine Research, Flødevigen Marine Research Station, His, Norway
[b]Present address: Department of Earth, Atmospheric, and Planetary Sciences, Massachusetts Institute of Technology, Cambridge, Massachusetts
[c]Present address: Department of Global Ecology, Carnegie Institution for Science, Stanford, California

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Temperature and nutrient availability are primary candidates for examining the effects of fluctuating conditions on ecosystem function and community composition as they both are fundamental environmental drivers with high dynamic ranges that govern metabolism, growth and population dynamics of most organisms, including marine plankton. The wide range of thermal tolerance that characterizes many planktonic species confers high adaptability (Thomas et al. 2012). Thus, it is unlikely that the forecasted increase in surface water temperature will exceed species' thermal limits, resulting in a complete elimination of ecological niches (Caron and Hutchins 2013; Franze and Lavrentyev 2014). The optimal temperature of growth of most planktonic species is often higher than in situ temperatures (Karentz and Smayda 1984). Thus, within a favorable range, temperature increases could positively affect metabolic rates in both autotrophic and heterotrophic plankton (Eppley 1972; Brown et al. 2004; Rose and Caron 2007; Chen 2022). Based on the theoretical Q10 model, the temperature dependence of growth predicts a doubling of the growth rate with every 10°C temperature increase (Q10 ≈ 2) (Eppley 1972). Nevertheless, laboratory studies on single phytoplankton species and strains reveal large inter- and intra-specific variability in growth responses to temperature (Thomas et al. 2012, 2016; Boyd et al. 2013, 2018; Godhe and Rynearson 2017; Barton and Yvon-Durocher 2019; Anderson and Rynearson 2020; Strock and Menden-Deuer 2021; Anderson et al. 2021). The few studies that have investigated the role of temperature in regulating herbivorous protist metabolic rates have found a similar high interspecific and intraspecific variability in the intensity and directionality of herbivore responses to changing temperature (Rose and Caron 2007; Franzè and Lavrentyev 2014; Menden-Deuer et al. 2018; Wang et al. 2019; Franzè and Menden-Deuer 2020).

This observed variability in herbivorous protist temperature dependence challenges theoretical predictions. In natural systems, a mismatch between observed and predicted temperature dependence of growth can reflect the combined effects of multiple drivers. For instance, environmental temperature affects water column stratification and nutrient input (Sarmiento et al. 2004) governing quality and quantity of nutrients availability to primary producers. This in turn drives phytoplankton competitive dynamics by affecting community composition and function (Litchman and Klausmeier 2008; Moran et al. 2018), and defining ecological niches and geographical distribution (Falkowski et al. 1998; Behrenfeld et al. 2005; Thomas et al. 2017; Rynearson et al. 2020; Anderson et al. 2022). Model studies have also predicted that by altering cellular nutrient uptake, increased environmental temperature could lead to both exacerbated nutrient limitation and higher metabolic rates, depending on the trophic status of the system (Serra-Pompei et al. 2019). Is has also been shown that nutrient limitation can reduce temperature sensitivity of phytoplankton productivity but also increase thermal sensitivity of growth by reducing the optimum temperature for growth (Thomas et al. 2017; Maranon et al. 2018). Moreover, it has been found that nutrient limitation could preclude thermal adaptation, leaving phytoplankton vulnerable to increasing temperatures (Aranguren-Gassis et al. 2019). The complexity of these interactions makes it unclear how direct temperature and nutrient effects on primary producers will affect their primary consumers, the herbivorous protists, which remove on average 2/3 of daily primary production globally, from polar to tropical oceans (Steinberg and Landry 2017).

Factoring in the ecosystem consequences of simultaneous shifts in temperature and nutrients on predator–prey interactions and trophic transfers multiplies the complexity of outcomes one can expect to observe in natural communities. Each added trophic level broadens the scope of potential ramifications of environmental stressors from direct effects to indirect and interactive effects. To our knowledge these interactions have been little studied experimentally.

Using a microcosm approach, we aimed to (1) quantify whether changes in temperature and nutrient concentrations have independent or synergistic effects on herbivorous protist grazing rates, (2) determine how shifts in the phytoplankton community would affect herbivorous protists grazing, and (3) measure how the synergistic or antagonistic effects of temperature and nutrient loading affect the magnitude of primary production removed and ultimately the net community production. Our study suggests that temperature has a direct effect on herbivorous protists and, depending on the trophic state of the system, temperature and nutrient availability exert a synergistic or antagonistic effect on plankton physiology with ramifications for trophic transfer rates and overall ecosystem functions.

Materials and methods

Seawater (SW) was collected from the Narragansett Bay (NB) Long-term Plankton Time Series site (41.57°N, 71.39°W; http://www.gso.uri.edu/phytoplankton/) on 20 March 2017 (Day 0, D0). Surface temperature and salinity were recorded using a 6920 multiparameter sonde (YSI Inc., Yellow Springs, Ohio). SW was filtered through a 200-μm mesh to eliminate macrozooplankton grazers, collected in 20-L acid-washed carboys, and immediately transported to the laboratory. At approximately the same time, additional 0.22 μm filtered SW (FSW) was collected from the University of Rhode Island Graduate School of Oceanography aquarium intake and stored to be used for dilution of the microcosms (*see* below). Temperature and salinity of the FSW (2.6°C; 30.5) were verified to be similar to the source SW (2.6°C, 29.3) with an additional 6920 multiparameter sonde (YSI Inc.). Glassware, plastic containers, and tubing used for the experimental set up were cleaned in 10% HCl, then rinsed with deionized water followed by SW.

The experiments presented here were conducted in parallel to Anderson et al. (2022) who investigated the impact of the

interactive effect of temperature and nutrients on phytoplankton community composition and physiology.

Microcosms set-up

The experimental set-up consisted of a nested design: seawater with plankton communities (< 200 μm) was used to set up long term (10 d) microcosm incubations at three temperatures and two nutrient concentrations to monitor the community response to temperature and nutrient manipulations in terms of species composition and abundance over the incubation period (Fig. 1a). The microcosms consisted of 12-L polypropylene carboys filled with 200 μm pre-screened SW and incubated for 10 d in temperature-controlled incubators (I-36LLVL Series, Percival Scientific) in duplicate at 2.6°C (collection temperature), −1°C, and 6°C. The temperature range selected for the incubations reflects spring surface temperature measured in Narraganset Bay (https://www.gso.uri.edu/phytoplankton/#Data). It should be noted that although the low temperature incubations began at −1°C they were adjusted to −0.5°C on Day 2 after some surface freezing occurred. The microcosms were incubated at a 12 : 12 light : dark cycle of cool white fluorescent lights at 115 μmol photons m^{-2} s^{-1}. To manipulate nutrient availability in the three temperature treatments, FSW either enriched with nutrients (nutrient amended; 32 μM nitrate, 2 μM phosphate, 32 μM silicate and f/27.6 concentration of vitamins and trace metals, final concentration) or enriched with only vitamins and trace metals (from here on referred to as nutrient unamended), depending on the treatment was added on Days 0, 3, and 6. The concentrations for nutrient amendments were

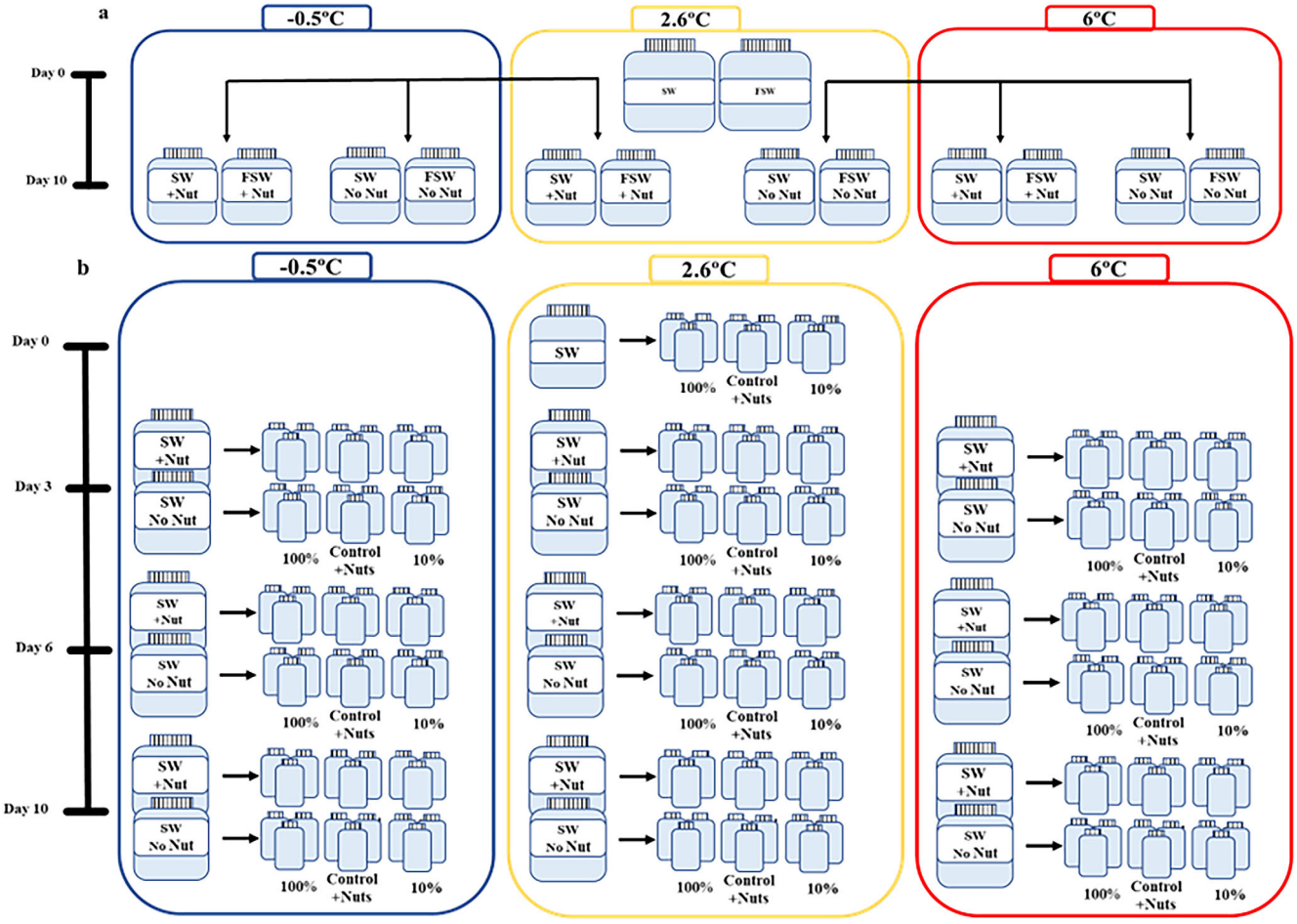

Fig. 1. Schematic representation of the experimental design. (**a**) Seawater (SW) and FSW were each split in six 12-L carboys. At each temperature, one set was amended with nutrients (*see* the text for concentrations) (+Nuts) and one only with vitamins and trace metals, without major nutrients (No Nuts). The incubation lasted for 10 d and on Days 0, 3, and 6 FSW with nutrients or vitamins without major nutrients was added to the treatments after water for the dilution experiments was collected in order to keep phytoplankton in exponential phase. (**b**) On Day 0 SW was used for the initial dilution experiment at in situ temperature (2.6°C). Subsequently on Days 3, 6, and 10 water from each microcosms was used to set up parallel dilution experiments. For each dilution experiment triplicates of 100% = SW and 10% = 10% SW + 90% FSW were incubated for 24 h. In addition, irrespectively from the microcosm nutrient treatment (+Nuts or No Nuts) a third set of bottles was incubated as nutrient control as requested from the dilution technique. Control + Nuts = 100% SW with addition of nutrients.

chosen to reflect the upper limit of concentrations recorded in Narragansett Bay as part of the long-term time series (https://www.gso.uri.edu/phytoplankton/#Data). The amendments on D3 and D6 were based on daily in vivo chlorophyll *a* (Chl *a*) autofluorescence measurements (*see* S2 in Anderson et al. 2022) in order to maintain phytoplankton in the exponential growth phase and Chl *a* concentration similar to in situ concentration measured on collection day. The FSW used to dilute the SW was kept in the same incubator as the experimental water in order to avoid temperature shock during the semi-continuous dilutions.

Dilution experiments

SW from each microcosm was used to assess phytoplankton growth and microzooplankton herbivory rates following the two-point modification of the dilution method (Landry and Hassett 1982) with a 100% and 10% SW dilution levels. Compared to a multipoint dilution series, the two-point modification provides statistically indistinguishable growth and grazing rate estimates for both linear and nonlinear feeding responses (Worden and Binder 2003; Strom and Fredrickson 2008; Chen 2015; Morison and Menden-Deuer 2017). The initial dilution experiment conducted on Day 0 (D0) was used to assess metabolic rates under in situ temperature and nutrient load. Then, on Day 3 (D3), Day 6 (D6), and Day 10 (D10) using water from each microcosm, 6 dilution experiments per day (one per each temperature and nutrient level) were conducted for a total of 19 dilution experiments in 10 d (Fig. 1b).

From each microcosm, after gentle rotation to mix the SW, SW was carefully siphoned into two sets of triplicate 500-mL polycarbonate bottles to prepare the 100% SW dilution levels. To satisfy one of the main assumptions of the dilution technique, that of unlimited phytoplankton growth, one of the 100% SW set, the Control + Nuts was amended with nutrients (32 μM nitrate, 2 μM phosphate, 32 μM silicate, and f/27.6 concentration of vitamins and trace metals, final concentration), irrespective of the nutrient amendment level of the source microcosms. The extra nutrient addition in the Control + Nuts allowed us to verify if phytoplankton growth rates were nutrient limited or not. The 10% treatments were prepared by mixing 90% FSW (prepared with water from each microcosm) with 10% SW in single carboys to minimize variation among replicates, before gentle siphoning into triplicate polycarbonate bottles. Experimental bottles were incubated for 24 h at −0.5°C, 2.6°C, and 6°C under a 12 : 12 light : dark cycle of cool white fluorescent lights at 115 μmol photons m^{-2} s^{-1}.

Triplicate subsamples were taken from the 100% SW stocks and after 24 h from each incubation bottle for Chl *a* and microscopy analysis. Chl *a* extraction and determination followed Graff and Rynearson (2011) and measurements were performed on a Turner Designs AU10 fluorometer. Plankton community enumeration and composition was performed on samples preserved in 2% acid Lugol's iodine final concentration (Menden-Deuer et al. 2001). Phytoplankton enumeration and dynamics are fully reported in Anderson et al. (2022). Here briefly, cells were enumerated using a Sedgewick–Rafter slide (1 mL volume) and a Nikon Eclipse E800 light microscope. Diatoms were identified to the lowest possible taxonomic level according to Tomas (1997) at a 200X–400X magnification. Microzooplankton were enumerated following the Utermöhl (1958) method settling between 2.5 and 15 mL. The entire surface area of the settling chamber was examined at 200X with a Nikon Diaphot 300 inverted microscope. Ciliates and dinoflagellates were identified and classified to the lowest possible taxonomic level consulting several taxonomic guides (Kofoid and Campbell 1929; Tomas 1997; Strüder-Kypke et al. 2002). A minimum of 100 cells each of ciliates and dinoflagellates > 15 μm were enumerated and sized with an eyepiece micrometer. Herbivorous protist biovolumes were calculated from their linear dimensions by approximating geometric shapes (Sun and Liu 2003) and converted to carbon (Menden-Deuer and Lessard 2000). Phytoplankton growth and herbivorous grazing rates were estimated from changes in total Chl *a* concentration over the 24 h incubation. The instantaneous phytoplankton growth rate (μ) depends on the assumption of unlimited, exponential growth and calculated following the equation: $\mu = 1/t \ln (N_t/N_0)$, where t is the incubation time in days and N_t and N_0 are the Chl *a* concentration at the beginning and at the end of the experiment. Herbivory rates due to microzooplankton grazing were estimated as the difference between μ measured in the diluted ($\mu_{10\%}$) and whole ($\mu_{100\%}$) SW sample $g = \mu_{10\%} - \mu_{100\%}$. The percentage of primary production consumed was calculated as $\%PP = (g/\mu_{10\%}) \times 100$. The potential prey field and thus the prey availability to the grazers, was assessed by dividing the abundance of the four dominant diatom genera (Anderson et al. 2022) by the abundance of herbivorous protists.

Statistical analyses

The direct and interactive effects of temperature and nutrient treatments on phytoplankton growth and protist herbivory rates were examined using a two-way ANOVA. Normality of data distributions and homoscedasticity of variance was ensured with a Shapiro–Wilk test. Bonferroni post hoc tests were conducted to correct for multiple comparisons and apply a conservative approach to identifying statistical significance. The equality of slopes was assessed through an analysis of covariance (ANCOVA). To detect the treatment effects on herbivorous protist species, abundance data were analyzed through a transformation-based principal component analysis (tb-PCA) applied to the Hellinger-transformed grazer species matrix using the vegan package (Oksanen 2018) in R. Nutrient and temperature vectors were fit to the ordination by applying envfit from the vegan package. In this analysis, vectors are fit using multiple regression and are oriented in the direction in which there is the greatest environmental change and to which they have maximal correlations with the PCA configuration. The significance of the vectors was then assessed using a permutation test.

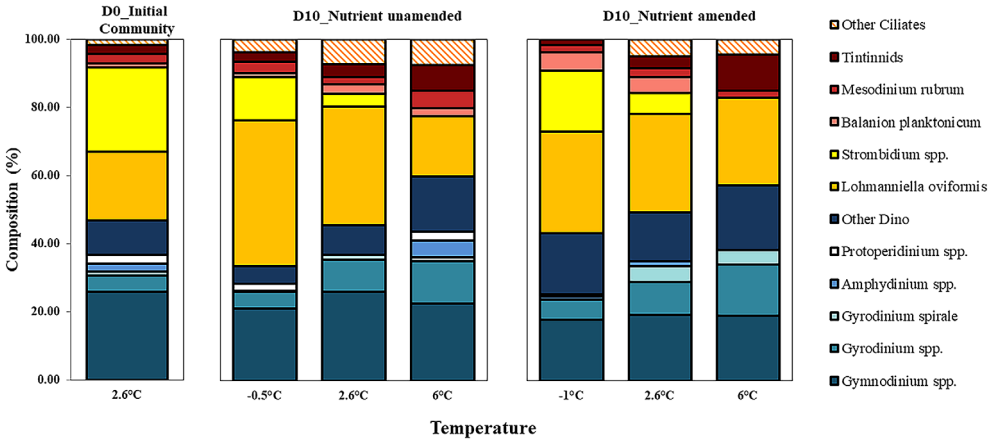

Fig. 2. Relative contribution of herbivorous protist taxa in the initial (D0) community and after 10 days (D10) of exposure to three temperature regimes under nutrient unamended or amended conditions. Species that contributed < 10% were combined in the "other" category for dinoflagellates and ciliates respectively. Dinoflagellates are identified in blue colors whereas yellow and reddish tones indicate ciliates.

All analyses were assigned statistical significance at $p < 0.05$ and were performed in R version 4.0.2 and Prism 7.

Results

Initial conditions

Seawater was characterized by low sea surface temperature (2.6°C) and high Chl *a* concentration (9.18 ± 0.37 μg L^{-1}). Microzooplankton abundance was $48{,}000 \pm 400$ cell L^{-1} with ciliates (53%) and dinoflagellates (47%) contributing equally to the initial population abundance (Fig. 2, D0). Similarly, dinoflagellate and ciliate contributed in equal measure to the total herbivorous biomass (31.44 and 37.5 μg C L^{-1}, respectively) with the largest contributors being ciliates and dinoflagellates larger than 20 μm in equivalent spherical diameter. Dinoflagellates were mostly represented by athecate species, with Gymnodiniales contributing 26% of total cell abundance. However, several thecate species including *Protoperidinium* spp. and *Amphydinium* spp. were present. Among ciliates, the numerically most abundant species were *Strombidium* spp. (24%) and *Lohmanniella oviformis* (20%) (Fig. 2, D0).

A detailed analysis of the phytoplankton species composition and their responses to temperature and nutrient manipulations is presented in the parallel study conducted by Anderson et al. (2022). Briefly, phytoplankton community was largely dominated by diatoms and particularly by four genera, *Chaetoceros*, *Leptocylindrus*, *Guinardia*, and *Skeletonema*, which together represented 95% of the entire population abundance. Microscopy showed that the majority of these cells (70%) were larger than 20 μm ESD, while most of the smaller phytoplankton cells were unidentified flagellates (< 2%).

The high Chl *a* concentration measured initially (D0) was paired with low growth (0.13 ± 0.03 d^{-1}) and grazing rates (0.06 ± 0.01 d^{-1}). The coupling between growth and grazing rates in this initial experiment resulted in the removal of 46% of primary production through consumption by microzooplankton.

Changes in community composition

Herbivorous protists' community composition, originally equally distributed between ciliates (53%) and dinoflagellates (47%), changed during the 10-d incubation in response to the changed environmental conditions (Fig. 2, D10). Ciliates and particularly *L. oviformis*, thrived in colder treatments where at the end of the 10-d incubation ciliates represented up to 66% of the predator population, while the dinoflagellate component represented a greater proportion in warmer temperature treatments (60%) (Fig. 2, D10). This differential temperature response was also suggested by the opposite alignment of ciliates and dinoflagellates with the temperature vector in the tb-PCA analysis (Fig. 3). Examining the overall response of herbivorous protist community composition to temperature and nutrient manipulation through tb-PCA analysis, we find that together the first two axes (tbPCA1 and tbPCA2) explained 66% of the total variance in the grazer community composition. Communities were significantly different between temperature treatments (permutation, $p = 0.006$), while the composition did not significantly vary with nutrient loads (permutation, $p = 0.300$; Figs. 2 and 3; Supporting Information Data S1).

However, smaller herbivores such as *S. epidemum* and *Gymnodinium* spp. showed a preference for lower nutrient conditions. Such small herbivores preferentially graze on bacteria, nano- and pico-size phytoplankton (Anderson and Rivkin 2001; Sherr and Sherr 2002), which are known to have an advantage under nutrient limiting conditions. It is also worthy of notice that some of the herbivorous species present in our community were directly observed preying upon large diatoms, showing active grazing on the diatom population (Fig. 4).

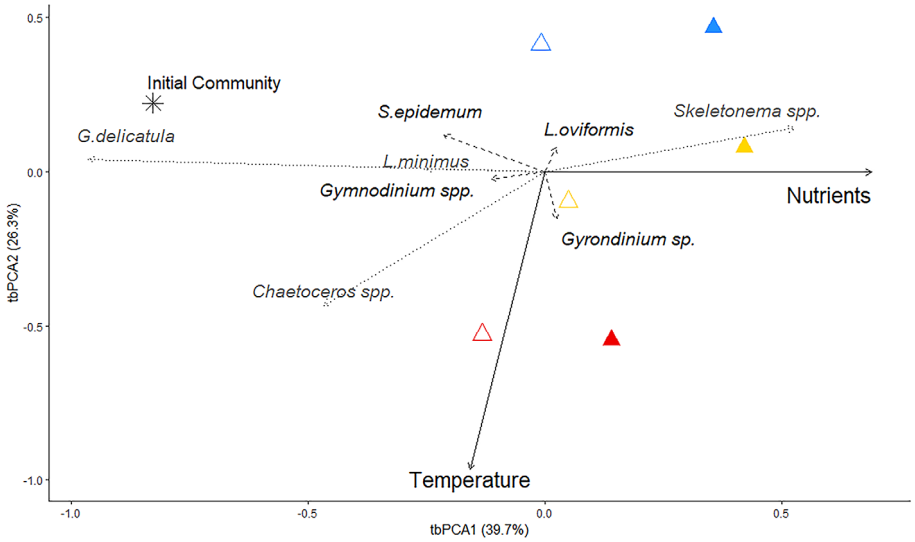

Fig. 3. Transformation-based principal component analysis (tb-PCA) of final (D10) herbivorous protist communities. Triangles represent overall herbivorous protist community composition for treatments at −0.5°C (blue), 2.6°C (yellow), and 6°C (red) under nutrient unamended (open triangles) or amended (filled triangles) conditions. Taxonomic information indicates the four herbivorous protists that presented the strongest response to experimental manipulation and the relative association of the four phytoplankton genera with herbivorous protist composition.

Among the four phytoplankton genera that dominate in our experimental samples, the diatom *Guinardia delicatula* was the only species whose abundance was significantly correlated (PCA, permutation $p = 0.008$) to herbivore community composition. Nevertheless, the alignment of *Skeletonema* with the nutrient vector suggest a positive association with the availability of nutrients, while *Leptocylindrus* thrived under nutrient limiting conditions (Fig. 3). The stronger response of these two genera to nutrient manipulation (Anderson et al. 2022) drove a shift in the prey field available to the herbivorous

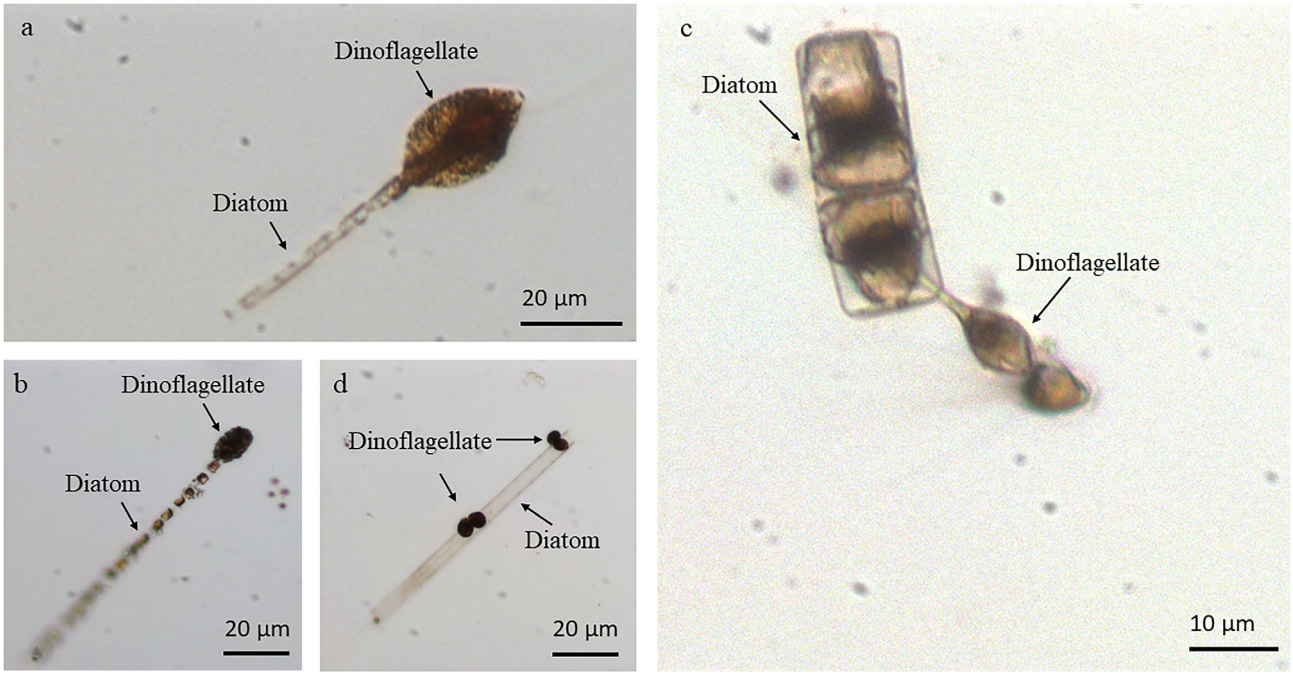

Fig. 4. Microscopy images capturing several dinoflagellate species engulfing (**a**,**b**) or pallium feeding (**c**,**d**) large diatom chains. The diatoms belong to the genus *Skeletonema* (**b**) or the species *Guinardia delicatula* (**c**). Prey identification was not possible in the other cases due to dinoflagellate consumption of key cellular structures.

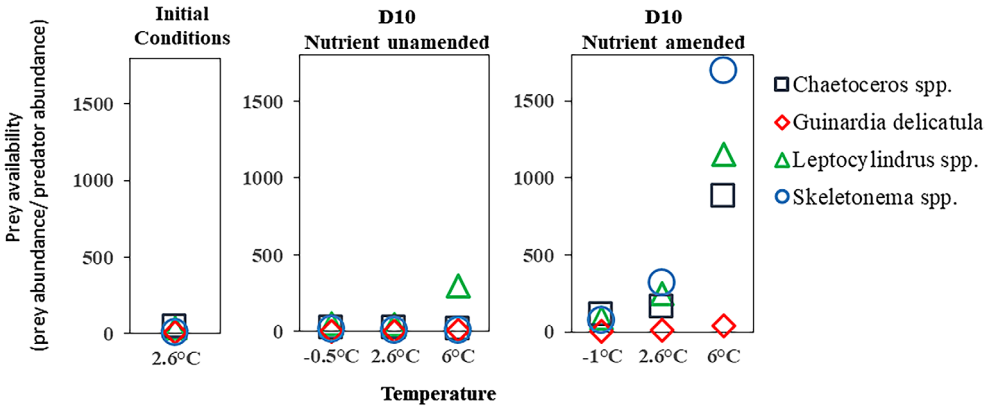

Fig. 5. Prey availability (prey abundance/predator abundance) based on microscopy counts for the initial (D0) and final (D10) plankton community after 10 days of exposure to nutrient unamended or amended conditions and under three temperature regimes.

grazers. Under in situ conditions *Leptocylindrus* and *Skeletonema* were available to the herbivorous grazers in a prey : predator abundance ratio of 40 : 1 and 13 : 1, respectively (Fig. 5, D0). These ratios remained unchanged throughout the incubations under nutrient unamended conditions except for *Leptocylindrus* whose increase in abundance in the warmest treatment translated in a sixfold (250 : 1) availability increase to the predators. On the other hand, under amended conditions both genera increased in abundance with increasing temperature. Thus, on D10 *Leptocylindrus* and *Skeletonema* availability to herbivorous protists was between 2- and 40-fold greater compared to the other genera (Fig. 5).

Plankton population dynamics

As would be expected by the initial high Chl *a* concentration, the phytoplankton community was nutrient limited. Irrespective of temperature treatment, phytoplankton growth rates were higher under nutrient amended vs. unamended conditions ($F_1 = 36.92$, two-way ANOVA, $p < 0.001$) (Fig. 6). A consistent trend of increase in phytoplankton growth over time was observed under nutrient amended conditions (Fig. 6, black dotted lines). Under unamended conditions, growth rates increased only at the lowest temperature, but not at the two higher temperature treatments.

At the lowest temperature there was no statistical difference (ANOVA, $p = 0.677$) between the phytoplankton growth rate measured in the 100% SW treatment and in the Control + Nuts suggesting that nutrient concentrations did not limit phytoplankton growth. This result is also reflected in the observed similarity of growth rates over time in the amended and unamended coldest microcosms ($F_{1,14} = 0.533$, ANCOVA, $p = 0.477$, Fig. 6a). It is worth noting that the initial exposure to − 1°C, which caused some limited and superficial freezing, might be the cause of the immediate, negative impact on phytoplankton growth. Despite readjusting the incubation temperature to −0.5°C, on D3 phytoplankton growth was either negative, indicating population decline, or 50% lower compared to phytoplankton growth rate measured on D0 (Fig. 6a). However, after this initial event, the phytoplankton community acclimated quite rapidly: after the initial temperature shock and despite the low absolute temperature, growth rates on D10 were about fourfold higher than on D3. At the two

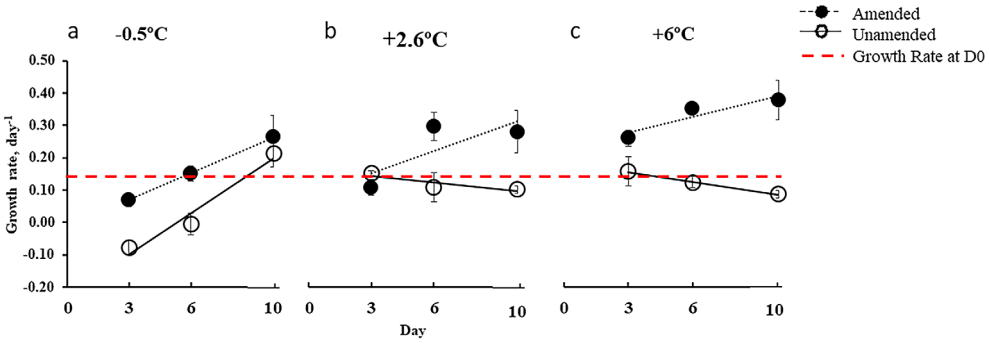

Fig. 6. Linear regression between phytoplankton growth rates and incubation days (D3, D6, and D10) at the three target temperatures (**a**–**c**) under unamended (open circles) and amended (filled circles) conditions. The red dotted line represents phytoplankton growth rates measured for the initial community (D0).

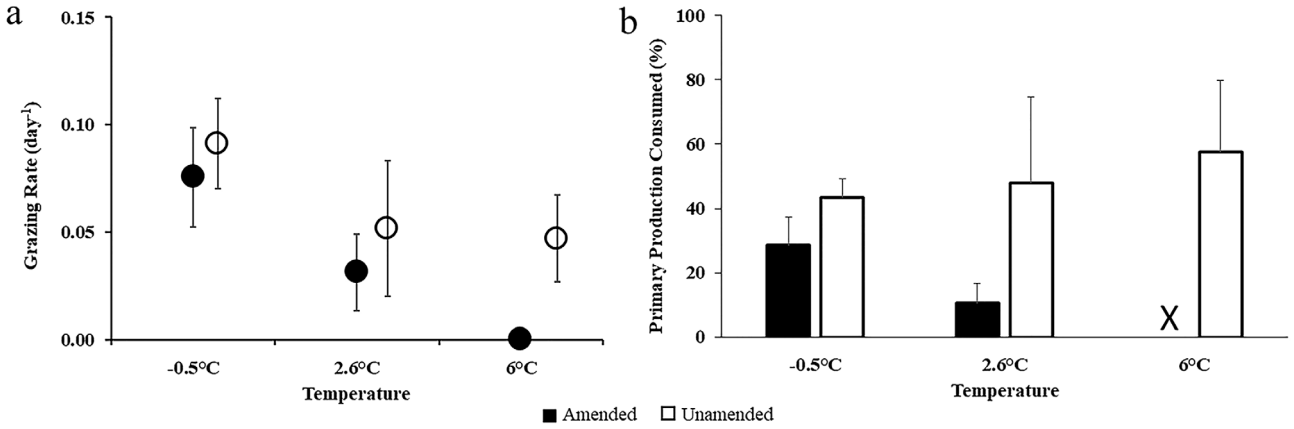

Fig. 7. (**a**) Final (D10) herbivorous protist grazing rates at each temperature under nutrient amended (filled circle) and unamended (open circles) conditions. (**b**) Percentage of primary production consumed under nutrient amended and unamended conditions and at three temperatures (−0.5°C, 2.6°C [in situ], and 6°C). Error bars represent standard deviation of triplicate measurements.

higher temperatures, nutrient limitation became evident as phytoplankton growth rates in the unamended treatments did not increase over time at the same rate as in the amended treatment. At 2.6°C, the in situ collection temperature, phytoplankton growth rates measured on D3 in the amended and unamended treatments were identical and not statistically different from the rate measured on D0 (Fig. 6b, red dotted line, $F_{2,6} = 0.31$; ANOVA, $p = 0.108$). However, phytoplankton growth rates changed over time depending on nutrient availability, and on D10 phytoplankton growth rate in the nutrient amended treatment was more than double the rate in the nutrient unamended treatment (Fig. 6b). A similar trend, although more pronounced, was observed in the highest temperature treatment (6°C). Under nutrient amended conditions, phytoplankton growth rate increased more rapidly and was already enhanced by D3; By D10 growth was more than threefold higher compared to the unamended treatment (Fig. 6c). In contrast, under nutrient unamended conditions, phytoplankton growth decreased over the 10-d incubation. The observed rates of decrease in growth over time in the unamended treatments at both 2.6°C and 6°C were not significantly different from each other (ANCOVA, $F_{1,14} = 0.63$, $p = 0.440$).

Throughout the experiments, herbivorous protist grazing rates were low, ranging between 0 and 0.09 ± 0.01 d^{-1} and, in general, higher in the unamended compared to the nutrient amended microcosms (Fig. 7a). Thus, the enhancement in phytoplankton growth and increase in prey availability through nutrient addition did not elicit a response in herbivorous grazing. The exposure to low temperature had no negative effect on protists' herbivory, in fact, the highest grazing rates were measured at the lowest temperature. On D10 grazing rates significantly decreased with increasing temperature irrespective of nutrient treatment (two-way ANOVA, $F_{2,8} = 20.10$, $p = 0.003$), while changes in nutrient availability had a marginally insignificant effect over time (two-way ANOVA, $F_{1,4} = 7.13$, $p = 0.056$). The difference between grazing rates in the amended and unamended treatment increased between onefold and fivefold with increasing temperature.

How much control herbivorous protists grazing exerted on phytoplankton populations under enhanced or depressed growing conditions was reflected in the amount of primary production consumed. With increasing temperature, microzooplankton grazing consumed a larger percentage of primary production in unamended conditions, while the opposite trend was observed under amended conditions where warming resulted in a decrease in primary production consumed (Fig. 7b). Thus, the highest fraction of primary production consumed ($58\% \pm 22\%$) was measured at high-temperature and low-nutrient concentration while at high-temperature and high-nutrient load no primary production was consumed by the herbivores. The higher fraction of primary production consumed in the high-temperature and low-nutrient treatment occurred because grazing rate decreased less (Fig. 7a) than phytoplankton growth rate (Fig. 6) under those conditions.

Discussion

This study examined the direct and indirect effects of changes in temperature and nutrient concentrations on phytoplankton growth and herbivorous protist grazing pressure and ultimately food web structure and function. We observed that (1) increasing temperature significantly depressed herbivorous protist grazing pressure on phytoplankton and that (2) nutrient loading had a lesser effect, likely indirectly due to nutrient induced changes in phytoplankton prey composition and population dynamics. The interaction of direct and indirect effects on both phytoplankton and herbivorous protists resulted in an increase in primary production consumed under nutrient unamended conditions and a decrease in consumption at warmer temperature and higher nutrient availability. At the ecosystem level, these changes to the food web

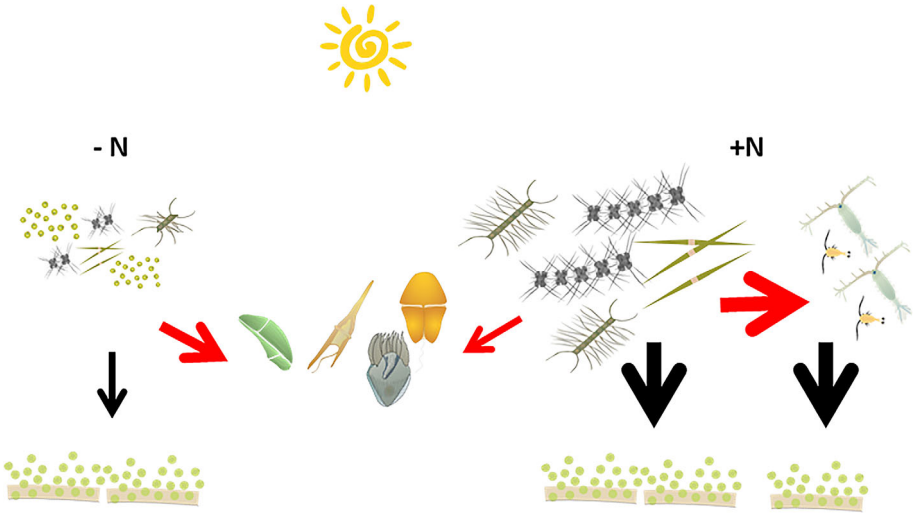

Fig. 8. Conceptual drawing representing possible responses in plankton population dynamics to nutrient inputs in a coastal marine ecosystem already thermally altered. *Left*: A warmer but nutrient regulated environment supports smaller phytoplankton with lower growth rates. Primary production is mostly consumed by herbivorous protists and enters the food web. *Right*: Warmer coastal waters with increased nutrient load support blooms of larger phytoplankton likely characterized by chemical and structural grazing deterrent mechanisms. Most of the primary production sinks out of the photic zone unconsumed fueling bacterial decomposition and the benthic food web or is consumed by larger mesozooplankton.

structure and function may determine the fate of primary and net community production. Under conditions of increased sea surface temperature, high-nutrient loads will favor blooms of fast-growing diatoms which might be subject to reduced grazing by microzooplankton due to a size mismatch and/or possible unpalatable characteristics. Under these circumstances protists would not consume most of the primary production which would instead be exported out of the pelagic zone or consumed by mesozooplankton. On the other hand, environments characterized by low or possibly controlled nutrient availability would support lower phytoplankton growth which could be better controlled by protist grazers strengthening food web interactions and trophic transfer (Fig. 8).

Temperature effects on phytoplankton and herbivorous protists

Previous studies have demonstrated that increasing temperature, within species upper thermal tolerance limit, triggers physiological responses in phytoplankton supporting higher growth rates (Eppley 1972; Brown et al. 2004; Kremer et al. 2017; Anderson et al. 2021). In line with these studies, through the dilution technique we observed temperature stimulation of phytoplankton growth under nonlimiting conditions (nutrient amended), showing that increased temperature had an overall positive effect on phytoplankton growth, total biomass, and species composition. A similar enhancement may have been in effect also for part of the dinoflagellates. Although species-specific growth rates were not assessed for herbivorous protists in this study, the observed community composition changes in close alinement with temperature, as identified by the tb-PCA analysis, suggests that dinoflagellates thrived in warmer waters. Mixotrophy, that is, the ability to combine autotrophic and heterotrophic modes of nutrition to satisfy energy requirements (Glibert and Mitra 2022), is prevalent among dinoflagellates and might be the mechanism behind the increased contribution observed.

This study was conducted in early spring and the conditions revealed that we sampled a polar plankton community, which is noteworthy because extensive coastal areas are in polar regions, and these coasts are subject both to warming and increased nutrient inputs. The plankton community studied here represents an early spring community collected at in situ temperature of 2.6°C. The collection site can be influenced by cold waters of Arctic origin during winter and early spring (Marrec et al. 2021), and a pan-Arctic nanodiatom (*Chaetoceros wighamii*) was isolated from these community incubations (Kling et al. 2021). Thus, it is safe to assume that this community had previously been subjected to subzero conditions and thus, was acclimated to low water temperatures (Franzè and Lavrentyev 2017; Lavrentyev et al. 2019). It is not surprising then that even in the coldest temperature treatment at the subzero °C phytoplankton quickly recovered from an initial temperature shock after few days of acclimation (Franzè and Menden-Deuer 2020) and, by the end of the incubation period phytoplankton grew at rates comparable to those observed at the higher in situ temperature of 2.6°C. Similarly, the small ciliate species *L. oviformis* that made up the majority of the microzooplankton community at −0.5°C, were clearly thriving and dividing at these low temperatures, as observed in several instances during our microscopy analysis.

Interactive effects and community implications

The independent influences of resource-dependency on species' growth, competition, and selection and of temperature-dependency on population, community, and ecosystem metabolisms are relatively well understood (Tilman 1982; Brown et al. 2004; Yvon-Durocher et al. 2010). However, it is the interactions and feedbacks between biological, chemical, and physical drivers that determine winners and losers in the rapidly changing surface ocean. The complexity of microbial systems makes it unlikely that the effect of multiple stressors results from the combined linear sum of the effects of single stressors. Investigations of interdependent components that govern ocean dynamics and their effects on lower trophic levels of the marine pelagic food webs reveal an intricate system of responses in which species interaction can exacerbate individual responses by increasing competition (Lewington-Pearce et al. 2019; Serra-Pompei et al. 2019). This highlights the necessity of simultaneously examining multiple factors and their interactive effects on multiple trophic levels when studying ecosystem responses.

The changes in grazing pressure observed here can only be explained by concurrently considering the interactive effects of temperature and nutrients manipulations on both phytoplankton and herbivorous protists. Despite the general increase in prey availability under warmer temperature the compositional shift and the rate of growth of phytoplankton driven by different nutrient loads, had a major role in determining the rate of herbivorous protist grazing. A shift toward less palatable species was observed specifically for the community investigated here, with the emergent dominance of large, chain-forming diatoms at higher temperatures and nutrient load (Anderson et al. 2022) which are less susceptible to herbivorous grazing due to their size and chemical and/or structural defense mechanisms (Van Donk et al. 2010). Factors that likely contribute to reduced grazing include *Chaetoceros* spines acting as a putative grazing deterrent (Pancic and Kiorboe 2018) for both microzooplankton and mesozooplankton predators. Moreover, although measurements of polyunsaturated aldehydes were outside the scope of this study, it is likely that some of the *Skeletonema* species present in our community (Anderson et al. 2022) released cytotoxic compounds reducing both growth and grazing rates of herbivorous protists (Lavrentyev et al. 2015; Franzè et al. 2017). The direct temperature effect on herbivorous protists species compositional shift that under nutrient amended conditions favored dinoflagellates and larger ciliates such as tintinnids and *Strombidium* sp. was not enough to compensate for the increase in growth and unpalatability of prey. In addition, a switch toward mixotrophy and a greater reliance on photosynthesis, would also explain why despite the increase in abundance of part of the grazers population, the grazing rate did not increase. However, mixotrophy was not measured in our experiments, so this hypothesis remains unresolved.

Based on our results, in coastal areas the synergistic effects of increased nutrient and temperature on planktonic communities would substantially lower the percentage of primary production consumed by herbivorous protists compared to prior studies (Franzé and Modigh 2013; Schmoker et al. 2013; Steinberg and Landry 2017). Therefore, we could expect that pulses of nutrients entering coastal systems already thermally altered, or vice versa, would lead to conditions where only a small amount of primary production would be entering the microbial food web, while the vast majority of production would remain available for export from the pelagic zone through sinking. Part of this available production could however be consumed by larger grazers (Fig. 8) even though comparative analyses show that mesozooplankton grazing represent only about ~ 25% of the total grazing impact (Campbell et al. 2009; Morison et al. 2020). If the synergistic effects of natural and anthropogenic driven changes create a mismatch between planktonic components preventing tight food web coupling and rapid flux of matter and energy, coastal communities could be facing significant ecosystem deterioration such as reduction of water quality, loss of biodiversity, decrease in natural resources and severe hypoxia (Rabalais et al. 2009) that may affect ecosystem services (i.e., fisheries and tourism) and potentially harm the economy (Moore et al. 2020).

If our results are applicable to other marine ecosystems, under nutrient unamended conditions an increase in temperature will amplify nutrient limitation, pushing phytoplankton communities to shift toward smaller cells with lower growth rates. For instance, the genus *Leptocylindrus* (and particularly the quite small *L. minimum*) dominated our unamended treatments due to their strong competitiveness under low nutrient concentrations (Anderson et al. 2022). Small phytoplankton cells, however, have shown to be more sensitive to changes in temperature than larger cells both experimentally and in meta-analyses (Maranon et al. 2018; Serra-Pompei et al. 2019). Predictions of decreased temperature optima for marine diatoms at low-nutrient concentrations suggest that species are more vulnerable to hot, low-nutrient conditions than previously thought (Thomas et al. 2017). These responses in phytoplankton community combined with the herbivorous protist community rearrangement allowed a better compositional match between predator and prey, creating a dynamic in which a significant amount of primary production (30%–43%) was processed through the microbial loop despite the overall low growth and grazing rates. Similar results were reported for a freshwater community where the responses to warming and nutrient addition strongly depended on the initial productivity status of the lakes (Schulhof et al. 2019). Thus, studies examining multi-factor, interaction effects on plankton populations and trophic dynamics demonstrably lead to very different and at times opposite conclusions than single factor studies.

It is noteworthy that, independently from the nutrient load, the highest grazing rates were measured at the lowest temperature (−0.5°C). This shows that plankton adapted to cold environments are not necessary physiologically depressed by subzero temperatures (Franze and Lavrentyev 2014; Franzè and

Lavrentyev 2017; Menden-Deuer et al. 2018; Franzè and Menden-Deuer 2020) and thus, play an essential role in channeling matter and energy toward higher trophic level also in high-latitude ecosystems, and temperate coasts in winter and spring.

Conclusions

The biological and trophic complexity of planktonic dynamics with intricate direct and indirect responses to co-varying drivers requires natural resource management to become increasingly holistic, with the focus shifting from individual species to whole ecosystems (Pikitch et al. 2004). There is substantial need to understand the relative importance of biotic interactions for the function and stability of ecosystems (Ives and Carpenter 2007). Given the fundamental role of plankton in aquatic systems, there is a need to better represent plankton population dynamics and variability in current food web models and other ecosystem model approaches (Lindemann et al. 2017). A first step in this direction is to characterize interactions within the plankton community while accounting for environmental conditions to identify the relative importance of direct environmental effects, density-dependent processes and trophic interactions. Following this approach, our study sheds light on complex ecosystem responses and suggests that under the scenario of ocean warming that we are experiencing, it is of paramount importance to be able to control and manage nutrient inputs in our coastal areas as nutrient loading directly impacts ecosystem production. The synergistic effect of increased temperature and high-nutrient load observed in this study suggests that a higher percentage of primary production could remain unconsumed, increasing concentrations of sinking organic matter or eutrophication and potentially deteriorating the health of coastal ecosystems.

Data availability statement

The data used in this publication have been submitted to BCO-DMO.

References

Anderson, M.R. and Rivkin, R.B., 2001. Seasonal patterns in grazing mortality of bacterioplankton in polar oceans: a bipolar comparison. Aquatic Microbial Ecology, **25**(2), pp.195-206.

Anderson, S. I., and T. A. Rynearson. 2020. Variability approaching the thermal limits can drive diatom community dynamics. Limnol. Oceanogr. **65**: 1961–1973. doi:10.1002/lno.11430

Anderson, S. I., A. D. Barton, S. Clayton, S. Dutkiewicz, and T. A. Rynearson. 2021. Marine phytoplankton functional types exhibit diverse responses to thermal change. Nat. Commun. **12**: 6413. doi:10.1038/s41467-021-26651-8

Anderson, S. I., and others. 2022. The interactive effects of temperature and nutrients on a spring phytoplankton community. Limnol. Oceanogr. **67**: 634–645. doi:10.1002/lno.12023

Aranguren-Gassis, M., C. T. Kremer, C. A. Klausmeier, and E. Litchman. 2019. Nitrogen limitation inhibits marine diatom adaptation to high temperatures. Ecol. Lett. **22**: 1860–1869. doi:10.1111/ele.13378

Barton, S., and G. Yvon-Durocher. 2019. Quantifying the temperature dependence of growth rate in marine phytoplankton within and across species. Limnol. Oceanogr. **64**: 2081–2091. doi:10.1002/lno.11170

Behrenfeld, M. J., E. Boss, D. A. Siegel, and D. M. Shea. 2005. Carbon-based ocean productivity and phytoplankton physiology from space. Global Biogeochem. Cycl. **19**: Gb1006. doi:10.1029/2004gb002299

Boyd, P. W., and others. 2013. Marine phytoplankton temperature versus growth responses from polar to tropical waters—Outcome of a scientific community-wide study. PLoS One **8**: e63091. doi:10.1371/journal.pone.0063091

Boyd, P. W., and others. 2018. Experimental strategies to assess the biological ramifications of multiple drivers of global ocean change—A review. Glob. Chang. Biol. **24**: 2239–2261. doi:10.1111/gcb.14102

Brown, J. H., J. F. Gillooly, A. P. Allen, V. M. Savage, and G. B. West. 2004. Toward a metabolic theory of ecology. Ecology **85**: 1771–1789. doi:10.1890/03-9000

Campbell, R. G., and others. 2009. Mesozooplankton prey preference and grazing impact in the western Arctic Ocean. Deep Sea Res. Part II **56**: 1274–1289. doi:10.1016/j.dsr2.2008.10.027

Caron, D. A., and D. A. Hutchins. 2013. The effects of changing climate on microzooplankton grazing and community structure: Drivers, predictions and knowledge gaps. J. Plankton Res. **35**: 235–252. doi:10.1093/plankt/fbs091

Chen, B. Z. 2015. Assessing the accuracy of the "two-point" dilution technique. Limnol. Oceanogr. Methods **13**: 521–526. doi:10.1002/lom3.10044

Chen, B. Z. 2022. Thermal diversity affects community responses to warming. Ecol. Model. **464**: 109846. doi:10.1016/j.ecolmodel.2021.109846

Eppley, R. W. 1972. Temperature and phytoplankton growth in the sea. Fish. Bull. **70**: 1063–1085.

Falkowski, P. G., R. T. Barber, and V. V. Smetacek. 1998. Biogeochemical controls and feedbacks on ocean primary production. Science **281**: 200–207. doi:10.1126/science.281.5374.200

Franzè, G., and P. J. Lavrentyev. 2014. Microzooplankton growth rates examined across a temperature gradient in the Barents Sea. PLoS One **9**: e86429. doi:10.1371/journal.pone.0086429

Franzè, G., and M. Modigh. 2013. Experimental evidence for internal predation in microzooplankton communities. Mar. Biol. **160**: 3103–3112. doi:10.1007/s00227-013-2298-1

Franzè, G., and P. J. Lavrentyev. 2017. Microbial food web structure and dynamics across a natural temperature

gradient in a productive polar shelf system. Mar. Ecol. Prog. Ser. **569**: 89–102. doi:10.3354/meps12072

Franzè, G., J. J. Pierson, D. K. Stoecker, and P. J. Lavrentyev. 2017. Diatom-produced allelochemicals trigger trophic cascades in the planktonic food web. Limnol. Oceanogr. **63**: 1093–1108. doi:10.1002/lno.10756

Franzè, G., and S. Menden-Deuer. 2020. Common temperature-growth dependency and acclimation response in three herbivorous protists. Mar. Ecol. Prog. Ser. **634**: 1–13. doi:10.3354/meps13200

Graff, J. R., and T. A. Rynearson. 2011. Extraction method influences the recovery of phytoplankton pigments from natural assemblages. Limnol. Oceanogr. Methods **9**: 129–139. doi:10.4319/lom.2011.9.129

Glibert, P. M., and A. Mitra. 2022. From webs, loops, shunts, and pumps to microbial multitasking: Evolving concepts of marine microbial ecology, the mixoplankton paradigm, and implications for a future ocean. Limnol. Oceanogr. **67**: 585–597. doi:10.1002/lno.12018

Godhe, A., and T. Rynearson. 2017. The role of intraspecific variation in the ecological and evolutionary success of diatoms in changing environments. Philos. Trans. R. Soc. **B 372**: 20160399. doi:10.1098/rstb.2016.0399

Hutchins, D. A., and F. Fu. 2017. Microorganisms and ocean global change. Nat. Microbiol. **2**: 17058. doi:10.1038/nmicrobiol.2017.58

Ives, A. R., and S. R. Carpenter. 2007. Stability and diversity of ecosystems. Science **317**: 58–62. doi:10.1126/science.1133258

Karentz, D., and T. Smayda. 1984. Temperature and seasonal occurrence patterns of 30 dominant phytoplankton species in Narragansett Bay over a 22-year period (1959–1980). Mar. Ecol. Prog. Ser. **18**: 277–293. doi:10.3354/MEPS018277

Kling, J. D., K. J. Kelly, S. Pei, T. A. Rynearson, and D. A. Hutchins. 2021. Irradiance modulates thermal niche in a previously undescribed low-light and cold-adapted nanodiatom. Limnol. Oceanogr. **66**: 2266–2277. doi:10.1002/lno.11752

Kofoid, C. A., and A. S. Campbell. 1929. A conspectus of the marine and freshwater ciliata belonging to the suborder Tintinnoinea, with description of new species principally from the Agassiz expedition to the Eastern Tropical Pacific 1904–1905. Univ. of California.

Kremer, C. T., M. K. Thomas, and E. Litchman. 2017. Temperature- and size-scaling of phytoplankton population growth rates: Reconciling the Eppley curve and the metabolic theory of ecology. Limnol. Oceanogr. **62**: 1658–1670. doi:10.1002/lno.10523

Landry, M. R., and R. P. Hassett. 1982. Estimating the grazing impact of marine micro-zooplankton. Mar. Biol. **67**: 283–288. doi:10.1007/Bf00397668

Lavrentyev, P. J., G. Franzè, J. J. Pierson, and D. K. Stoecker. 2015. The effect of dissolved polyunsaturated aldehydes on microzooplankton growth rates in the Chesapeake Bay and Atlantic coastal waters. Mar. Drugs **13**: 2834–2856. doi:10.3390/md13052834

Lavrentyev, P. J., G. Franzè, and F. B. Moore. 2019. Microzooplankton distribution and dynamics in the Eastern Fram Strait and the Arctic Ocean in May and August 2014. Front. Mar. Sci. **6**: 264. doi:10.3389/fmars.2019.00264

Lewington-Pearce, L., A. Narwani, M. K. Thomas, C. T. Kremer, H. Vogler, and P. Kratina. 2019. Temperature-dependence of minimum resource requirements alters competitive hierarchies in phytoplankton. Oikos **128**: 1194–1205. doi:10.1111/oik.06060

Lindemann, C., D. L. Aksnes, K. J. Flynn, and S. Menden-Deuer. 2017. Editorial: Modeling the plankton-enhancing the integration of biological knowledge and mechanistic understanding. Front. Mar. Sci. **4:** 358. doi:10.3389/fmars.2017.00358

Litchman, E., and C. A. Klausmeier. 2008. Trait-based community ecology of phytoplankton. Annu. Rev. Ecol. Evol. Syst. **39**: 615–639. doi:10.1146/annurev.ecolsys.39.110707.173549

Maranon, E., M. P. Lorenzo, P. Cermeno, and B. Mourino-Carballido. 2018. Nutrient limitation suppresses the temperature dependence of phytoplankton metabolic rates. ISME J. **12**: 1836–1845. doi:10.1038/s41396-018-0105-1

Marrec, P., H. McNair, G. Franzè, F. Morison, J. P. Strock, and S. Menden-Deuer. 2021. Seasonal variability in planktonic food web structure and function of the Northeast U.S. Shelf. Limnol. Oceanogr. **66**: 1440–1458. doi:10.1002/lno.11696

Menden-Deuer, S., C. Lawrence, and G. Franzè. 2018. Herbivorous protist growth and grazing rates at in situ and artificially elevated temperatures during an Arctic phytoplankton spring bloom. PeerJ **6**: e5264. doi:10.7717/peerj.5264

Menden-Deuer, S., and E. J. Lessard. 2000. Carbon to volume relationships for dinoflagellates, diatoms, and other protist plankton. Limnol. Oceanogr. **45**: 569–579. doi:10.4319/lo.2000.45.3.0569

Menden-Deuer, S., E. J. Lessard, and J. Satterberg. 2001. Effect of preservation on dinoflagellate and diatom cell volume and consequences for carbon biomass predictions. Mar. Ecol. Prog. Ser. **222**: 41–50. doi:10.3354/meps222041

Moore, S. K., and others. 2020. Harmful algal blooms and coastal communities: Socioeconomic impacts and actions taken to cope with the 2015 U.S. West Coast domoic acid event. Harmful Algae **96**: 101799. doi:10.1016/j.hal.2020.101799

Moran, X. A. G., A. Calvo-Diaz, N. Arandia-Gorostidi, and T. M. Huete-Stauffer. 2018. Temperature sensitivities of microbial plankton net growth rates are seasonally coherent and linked to nutrient availability. Environ. Microbiol. **20**: 3798–3810. doi:10.1111/1462-2920.14393

Morison, F., and S. Menden-Deuer. 2017. Doing more with less? Balancing sampling resolution and effort in

measurements of protistan growth and grazing-rates. Limnol. Oceanogr. Methods **15**: 794–809. doi:10.1002/lom3.10200

Morison, F., G. Franzè, E. Harvey, and S. Menden-Deuer. 2020. Light fluctuations are key in modulating plankton trophic dynamics and their impact on primary production. Limnol. Oceanogr. Lett. **5**: 346–353. doi:10.1002/lol2.10156

Oksanen, J., and others. 2018. Vegan: Community ecology package. R package version 2.5-6. Available from https://CRAN.R-project.org/package=vegan

Pancic, M., and T. Kiorboe. 2018. Phytoplankton defence mechanisms: Traits and trade-offs. Biol. Rev. Camb. Philos. Soc. **93**: 1269–1303. doi:10.1111/brv.12395

Pikitch, E. K., and others. 2004. Ecology. Ecosystem-based fishery management. Science **305**: 346–347. doi:10.1126/science.1098222

Rabalais, N. N., R. E. Turner, R. J. Diaz, and D. Justic. 2009. Global change and eutrophication of coastal waters. ICES J. Mar. Sci. **66**: 1528–1537. doi:10.1093/icesjms/fsp047

Rose, J. M., and D. A. Caron. 2007. Does low temperature constrain the growth rates of heterotrophic protists? Evidence and implications for algal blooms in cold waters. Limnol. Oceanogr. **52**: 886–895. doi:10.4319/lo.2007.52.2.0886

Rynearson, T. A., S. A. Flickinger, and D. N. Fontaine. 2020. Metabarcoding reveals temporal patterns of community composition and realized thermal niches of *Thalassiosira* spp. (Bacillariophyceae) from the Narragansett Bay long-term plankton time series. Biology **9**: 19. doi:10.3390/biology9010019

Sarmiento, J. L., and others. 2004. Response of ocean ecosystems to climate warming. Global Biogeochem. Cycles **18**: Gb3003. doi:10.1029/2003gb002134

Schmoker, C., S. Hernandez-Leon, and A. Calbet. 2013. Microzooplankton grazing in the oceans: Impacts, data variability, knowledge gaps and future directions. J. Plankton Res. **35**: 691–706. doi:10.1093/plankt/fbt023

Schulhof, M. A., J. B. Shurin, S. A. J. Declerck, and D. B. Van de Waal. 2019. Phytoplankton growth and stoichiometric responses to warming, nutrient addition and grazing depend on lake productivity and cell size. Glob. Chang. Biol. **25**: 2751–2762. doi:10.1111/gcb.14660

Serra-Pompei, C., G. I. Hagstrom, A. W. Visser, and K. H. Andersen. 2019. Resource limitation determines temperature response of unicellular plankton communities. Limnol. Oceanogr. **64**: 1627–1640. doi:10.1002/lno.11140

Sherr, E. B., and B. F. Sherr. 2002. Significance of predation by protists in aquatic microbial food webs. Antonie van Leeuwenhoek **81**: 293-308.

Steinberg, D. K., and M. R. Landry. 2017. Zooplankton and the ocean carbon cycle. Ann. Rev. Mar. Sci. **9**: 413–444. doi:10.1146/annurev-marine-010814-015924

Strock, J. P., and S. Menden-Deuer. 2021. Temperature acclimation alters phytoplankton growth and production rates. Limnol. Oceanogr. **66**: 740–752. doi:10.1002/lno.11637

Strom, S. L., and K. A. Fredrickson. 2008. Intense stratification leads to phytoplankton nutrient limitation and reduced microzooplankton grazing in the southeastern Bering Sea. Deep Sea Res. Part II **55**: 1761–1774. doi:10.1016/j.dsr2.2008.04.008

Strüder-Kypke, M. C., S. D. Agatha, J. Warwick, and D. J. S. Montagnes. 2002. The user-friendly key to coastal planktonic ciliates.

Sun, J., and D. Y. Liu. 2003. Geometric models for calculating cell biovolume and surface area for phytoplankton. J. Plankton Res. **25**: 1331–1346. doi:10.1093/plankt/fbg096

Thomas, M. K., C. T. Kremer, C. A. Klausmeier, and E. Litchman. 2012. A global pattern of thermal adaptation in marine phytoplankton. Science **338**: 1085–1088. doi:10.1126/science.1224836

Thomas, M. K., C. T. Kremer, and E. Litchman. 2016. Environment and evolutionary history determine the global biogeography of phytoplankton temperature traits. Glob. Ecol. Biogeogr. **25**: 75–86. doi:10.1111/geb.12387

Thomas, M. K., and others. 2017. Temperature-nutrient interactions exacerbate sensitivity to warming in phytoplankton. Glob. Chang. Biol. **23**: 3269–3280. doi:10.1111/gcb.13641

Tilman, D. 1982. Resource competition and community structure. Princeton Press.

Tomas, C. K. 1997. Identifying marine phytoplankton. Academic Press.

Utermöhl, H. 1958. Zur Ver vollkommung der quantitativen phytoplankton-methodik. Mitteil. Int. Ver. Theor. Amgew. Limnol. **9**: 39.

Van Donk, E., A. Ianora, and M. Vos. 2010. Induced defences in marine and freshwater phytoplankton: A review. Hydrobiologia **668**: 3–19. doi:10.1007/s10750-010-0395-4

Wang, Q., Z. Lyu, S. Omar, S. Cornell, Z. Yang, and D. J. S. Montagnes. 2019. Predicting temperature impacts on aquatic productivity: Questioning the metabolic theory of ecology's "canonical" activation energies. Limnol. Oceanogr. **64**: 1172–1185. doi:10.1002/lno.11105

Worden, A. Z., and B. J. Binder. 2003. Application of dilution experiments for measuring growth and mortality rates among *Prochlorococcus* and *Synechococcus* populations in oligotrophic environments. Aquat. Microb. Ecol. **30**: 159–174. doi:10.3354/ame030159

Yvon-Durocher, G., J. I. Jones, M. Trimmer, G. Woodward, and J. M. Montoya. 2010. Warming alters the metabolic balance of ecosystems. Philos. Trans. R. Soc. B **365**: 2117–2126. doi:10.1098/rstb.2010.0038

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Conflict of interest statement

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