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RESEARCH ARTICLE

Phytoplankton diversity affects biomass and energy production differently during community development

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Abstract

1. Biodiversity determines the productivity and stability of ecosystems but some aspects of biodiversity–ecosystem functioning relationships remain poorly resolved. One key uncertainty is the inter-relationship between biodiversity, energy and biomass production as communities develop over time. Energy production drives biomass accumulation but the ratio of the two processes can change during community development. How biodiversity affects these temporal patterns remains unknown.
2. We empirically assessed how species diversity mediates the rates of increase and maximum values of biomass and net energy production in experimental phytoplankton communities over 10 days in the laboratory. We used five phytoplankton species to assemble three levels of diversity (monocultures, bicultures and communities) and we quantify their changes in biomass production and energy fluxes (energy produced by photosynthesis, consumed by metabolism, and net energy production as their difference) as the cultures move from a low density, low competition system to a high density, high competition system.
3. We find that species diversity affects both biomass and energy fluxes but in different ways. Diverse communities produce net energy and biomass at faster rates, reaching greater maximum biomass but with no difference in maximum net energy production. Bounds on net energy production seem stronger than those on biomass because competition limits energy fluxes as biomass accumulates over time.
4. In summary, diversity initially enhances productivity by diffusing competitive interactions but metabolic density dependence reduces these positive effects as biomass accumulates in older communities. By showing how biodiversity affects both biomass and energy fluxes during community development, our results demonstrate a mechanism that underlies positive biodiversity effects and offer a framework for comparing biodiversity effects across systems at different stages of development and disturbance regimes.

KEYWORDS
biodiversity–ecosystem functioning, competition, ecosystem functioning, metabolism, species turnover

1 | INTRODUCTION

The global loss of species is a concern because biodiversity enhances biological production and resistance to disturbance (Cardinale et al., 2012; Isbell et al., 2015). Diverse communities are often more productive because functional complementarity boosts resource use and because they are more likely to contain very productive species (complementarity and selection effects respectively; Cardinale et al., 2007; Loreau & Hector, 2001; Weisser et al., 2017). However, some aspects of biodiversity–ecosystem functioning relationships remain unclear (Loreau et al., 2012), particularly when extrapolating across ecosystem types and larger scales (Brose & Hillebrand, 2016; Gonzalez et al., 2020). One way to improve our understanding of biodiversity–ecosystem functioning (BEF) relationships is to examine lower-level mechanisms and identify specific biological interactions that lead to overyielding (e.g. N-fixing legumes in grasslands; Hooper et al., 2005; Tilman et al., 1997). A complementary but less explored approach is to consider how fundamental ecosystem processes, such as energy fluxes, drive and constrain BEF relationships (Barnes et al., 2018; Buzhdygan et al., 2020; Loreau, 2010)—which is the approach we take here.

Assessments of energy fluxes are a central component of ecosystem ecology (Lindeman, 1942; MacArthur, 1969). During ecosystem development, energy production fuels biomass growth but the ratio of these two processes changes over time: older communities often sustain more biomass per unit energy flow (Boit & Gaedke, 2014; Odum, 1969). Whether initial levels of biodiversity affect the relationship between energy and biomass production remains, however, unknown because of the lack of overlap between fields: succession theory overlooks biodiversity effects and BEF studies rarely assess how energy fluxes (and their relationship to biomass production) change during community development (Buzhdygan et al., 2020). The lack of integration between theories is problematic because disturbances often disrupt community development and reduce biodiversity simultaneously. Since disturbances are widespread, comparisons across natural systems are difficult without a clear understanding of how biodiversity affects production during community development.

The main focus of BEF research has been on resource uptake, which is the first step in the movement of energy across trophic levels. However, a greater uptake of resources does not necessarily boost production which also depends on how those resources are converted into biomass (Hodapp et al., 2019). Critically, resource uptake and metabolic expenditure can respond differently to changes in population densities (Ghedini et al., 2017; Malerba et al., 2017) and the efficiency of these processes varies as competition intensifies over time (Gatto, 1990; Ghedini, Loreau, et al., 2020; MacArthur, 1969). Therefore, biomass production should depend on the sensitivity of energy fluxes to competition, and the effects of biodiversity on this relationship.

The effects of biodiversity on ecosystem functioning should vary over succession for most systems because changes in species dominance and competition intensity during succession affect complementarity and selection effects (Armitage, 2016; Fargione et al., 2007). Yet, the temporal dynamics of BEF relationships have been assessed by few studies and typically only on biomass (but see Armitage, 2016 for respiration). These studies include natural succession in the field (Lasky et al., 2014) and experimental assessments on fixed pools of species (Weis et al., 2007) or species pools isolated at different time points of community development (Armitage, 2016; Wacker et al., 2009). These studies consistently show a peak in positive BEF effects in early or intermediate stages of development and a decline in older communities. The effects of biodiversity on biomass, therefore, appear predictable during succession, with or without species turnover (i.e. the arrival of new species at different time points). Whether biodiversity has similar effects on energy fluxes and whether these effects explain patterns of biomass production remain to be tested.

We use a simple but tractable experimental system of marine phytoplankton to study the inter-relationship between biodiversity, energy fluxes and biomass production during community development in the laboratory. Specifically, we test how species diversity affects biomass production (biovolume) and fluxes of energy (production through photosynthesis, consumption through metabolism and net energy production as their difference over a 24-hr period) as competition intensifies over time—from the point of initial colonisation, when species densities are low and competition is weak, to older communities dominated by strong competitive interactions (similarly to Weis et al., 2007). Our experiment does not mimic natural succession because we do not account for species immigration at different points in time (e.g. Armitage, 2016). Nonetheless, our results can provide valuable insights into successional processes because changes in the intensity of competition are a key component of succession.

Marine phytoplankton are the major primary producers in aquatic ecosystems, driving 50% of carbon uptake and primary production (Litchman et al., 2007). It is thus fundamental to understand how phytoplankton diversity affects these critical functions at different successional stages. In our assessment, we use five cosmopolitan species to assemble three levels of diversity (monocultures, all combinations of species pairs and communities with all five species) using a substitutive design so that each culture had an equal total biovolume and equal biovolume of each species at the start of the experiment. The species we use span two orders of magnitude in size and four phytoplankton groups with different photosynthetic pigments, encompassing a range of growth and resource use strategies that mediate competition for nutrients and light (Burson et al., 2018; Litchman et al., 2007). We follow the dynamics of these species and measure changes in biomass and net energy production at each level of diversity over 10 days (~10 generations) to quantify differences in their maximum rates of increase (r_{max}) and maximum values (K). We then interpret these results in light of the strength of competition and the production of energy as biomass grows.

2 | MATERIALS AND METHODS

2.1 | Species and diversity treatments

We used five cosmopolitan species of marine phytoplankton to assemble 16 species combinations over three levels of species diversity (one, two and five species—hereafter called monocultures, pairs and communities). Each monoculture and species pair were replicated three times and the communities with all five species were replicated five times ($N = 50$ cultures). The species were: *Amphidinium carterae* (CS-740, Dinoflagellata; initial average cell size = 309.9 ± 15.6 μm^3), *Tetraselmis* sp. (CS-91, Chlorophyta; 168.3 ± 7.8 μm^3), *Dunaliella tertiolecta* (CS-14, Chlorophyta; 110.9 ± 5.3 μm^3), *Tisochrysis lutea* (CS-177, Haptophyta; 24.3 ± 1.3 μm^3) and *Synechococcus* sp. (CS-94, Cyanobacteria; 5.4 ± 0.3 μm^3). These species span a range of cell sizes, growth strategies and pigments, and can coexist for over 6 weeks under laboratory conditions (Table S1; Ghedini, Loreau, et al., 2020). Strains were obtained from the Australian National Algae Culture Collection. Each species was cultured individually as a mother culture in 2 L glass bottles for 2 months before the experiment using standard *f*/2 enriched seawater medium (Nitrogen = 882 μM, Phosphorus = 36.2 μM) designed for growing coastal marine algae, prepared with 0.45 μm filtered seawater and autoclaved (Guillard & Ryther, 1962; Jeffrey & LeRoi, 1997). During the experiment, which lasted 10 days, each culture was grown in a 200 ml clear culture flask filled to 100 ml and kept in a temperature-controlled room at 22 ± 1°C. Cultures were grown on a 14–10 hr light–dark cycle under non-saturating irradiance levels (115 ± 5 $\mu mol\ m^{-2}\ s^{-1}$; Six et al., 2004; Yang et al., 2020). Flasks were shaken and randomly rearranged on the shelves every day. Nutrients were added daily by replacing 10% of medium from each flask (used for sampling) with fresh *f*/2 medium (dilution 0.1/day).

We adopted a substitutive design so, at the start of the experiment (2 December 2019), we inoculated the flasks with the same total biovolume of 5.68×10^8 μm^3 and equal biovolume of each species within treatments. To calculate the initial biovolume of each species, we determined the cell abundance (cells/μl) and average cell volume (μm^3, 50 cells per species) of each mother culture with an Olympus light microscope at 400× magnification from 2 × 10 μl samples stained with 1% Lugol's iodine. Cell abundances were determined with a Neubauer counting chamber (ProSciTech, Australia), while cell sizes were measured from photos using ImageJ and Fiji (version 2.0; Schindelin et al., 2012), assigning to each species an approximate geometric shape (Table S1; Hillebrand et al., 1999). We calculated the biovolume of each species as the product of their cell size and abundance ($\mu m^3/\mu l$) to determine the species with limiting biovolume. We chose a priori to add 20 ml of this species to its monoculture (20% of total volume) and back calculated the volumes of the other species. Each flask was then filled with standard *f*/2 medium to 100 ml. We then sampled 1 ml from one replicate per treatment of monocultures and species pairs and all five replicates of communities to check species biovolumes relative to the target (day_0).

2.2 | Data collection

We quantified biovolume production and energy fluxes for 10 days, corresponding to approximately 10 generations (Laws, 2013). Data were collected each day for the first five days and at alternate days afterwards for a total of eight sampling times (day 0, 1, 2, 3, 4, 6, 8 and 10). One millilitre sample from each replicate culture was fixed with Lugol's solution (1%) to quantify the abundance and cell size of each species as described above. The total biovolume of the culture was calculated as the sum of individual species biovolumes for pairs and communities. On the same day, we measured oxygen evolution rates (net photosynthesis, post-illumination and dark metabolism) from a 5 ml sample of each culture. Oxygen evolution rates were calculated from change in percentage oxygen saturation using 24-channel sensor dish readers (SDR; PreSens Precision Sensing GmbH) following established protocols (Ghedini, Loreau, et al., 2020; Malerba et al., 2017). Briefly, sensors were calibrated with 0% and 100% air saturation before the experiment. Net photosynthesis was measured at the same light intensity at which the cultures were grown for 1 hr (or until reached 250% air saturation, the max. measurable), followed by 1 hr in the dark to measure respiration rates. Twelve blanks were filled with the supernatant of centrifuged samples (spun at 2,500 rpm for 10 min to separate the algae from the supernatant) to correct for background microbial activity since cultures were not axenic. Prior to measurements, all samples were spiked with 50 μl of sodium bicarbonate stock for a final concentration of 2 mM sodium bicarbonate to avoid carbon limitation.

The rate of photosynthesis or metabolism of the whole sample (VO_2; units $\mu mol\ O_2/min$) was measured as $VO_2 = 1 \times ((m_a - m_b)/100 \times V\beta O_2)$ following (White et al., 2011), where m_a is the rate of change of O_2 saturation in each sample (min^{-1}), m_b is the mean O_2 saturation across all blanks (min^{-1}), V is the sample volume (0.005 L) and $V\beta O_2$ is the oxygen capacity of air-saturated seawater at 20°C and 35 ppt salinity (225 $\mu mol\ O_2/L$). The first 3 minutes of measurements were discarded for all samples and post-illumination metabolism was calculated over the first 10 min of the dark measurement as this faster oxygen consumption indicated enhanced post-illumination metabolic rates (Beardall et al., 1994). Finally, photosynthesis and metabolism ($\mu mol\ O_2/min$) were converted to calorific energy (J/min) using the conversion factor of 0.512 $J/\mu mol\ O_2$ (Williams & Laurens, 2010) to estimate energy production and energy consumption respectively. As a last step, we calculated the net energy production of the whole sample over a 24-hr period (J/day) as 14 hr of energy produced through net photosynthesis minus 0.25 hr of post-illumination metabolism and 9.75 hr of dark metabolism.

2.3 | Biodiversity effects on biovolume

We calculated complementarity (CE), selection (SE) and net biodiversity effects (NBEs) on biovolume for species pairs and communities following (Loreau & Hector, 2001). The deviation from the expected biovolume yield of a species *i* in the mixture at each point in time (day$_x$) was calculated as $\Delta \mathrm{RY}i = \left(\frac{Yi}{Mi}\right) - \left(\frac{Yi_{\mathrm{in}}}{\mathrm{tot}Y_{\mathrm{in}}}\right)$ where Y_i is the observed yield of species *i* in the mixture and M_i the monoculture yield of that species at day$_x$, and Yi_{in} and $\mathrm{tot}Y_{\mathrm{in}}$ are the initial yield of species *i* and total yield of the mixture respectively (i.e. initial proportion at day$_0$). Complementarity effects were calculated as $N \times \overline{\Delta \mathrm{RY}} \times \overline{M}$, where N is the number of species in the mixture, $\overline{\Delta \mathrm{RY}}$ is the average change in relative yield for all species in the mixture and $\overline{M}$ is the average monoculture biomass across all species in the mixture at each point in time. Selection effects were calculated as $N \times \mathrm{Cov}(\Delta \mathrm{RY}, M)$, that is the covariance between the monoculture yield of a species (M) and its change in relative yield in the mixture (ΔRY), multiplied by the number of species in the mixture. The net biodiversity effect is the sum of CE and SE. Finally, we plot NBE, CE and SE over time including the mean and 95% confidence intervals obtained with a nonparametric bootstrap without assuming normality (function mean_cl_boot in Hmisc r package). We used changes in biovolume of each species to calculate Pielou's measure of species evenness (*J*').

2.4 | Estimates of competitive interactions

To estimate the strength of intra- and interspecific competition, we estimated competition coefficients for each pair of species *ij* by fitting Lotka–Volterra competition models to the time series of cell density counts. For each species pair, we combined the cell counts of species *i* (or *j*) in monoculture ($n = 3$) with those of the pair *ij* ($n = 3$ for each competitor species *j*) to assess the effects of species *j* on species *i* across a wide range of densities (Inouye, 2001). Instead of modelling the Lotka–Volterra model with a carrying capacity term, we explicitly modelled the absolute intra- and interspecific competition coefficients for each pairs of species (Adler et al., 2018):

$$\frac{dN_i}{dt} = r_i N_i \left(1 - \alpha_{ii} N_i - \alpha_{ij} N_j\right), \tag{1}$$

$$\frac{dN_j}{dt} = r_j N_j \left(1 - \alpha_{jj} N_j - \alpha_{ji} N_i\right), \tag{2}$$

where N_i (N_j) is the cell density (cells/μl) and r_i (r_j) is the intrinsic growth rate of species *i* (*j*). The competition coefficients α represent the effect of an increase in conspecific (α_{ii} or α_{jj}) and heterospecific density on population growth (α_{ij} for the effect of species *j* on *i*, and, *vice versa*, α_{ji}). Note that the α_{ij} obtained in (1) is equivalent to α'_{ij}/K_i where α'_{ij} is the relative competition coefficient estimated by modelling the carrying capacity K_i of species *i* and assuming an intraspecific competition coefficient of 1. We then used all four competition coefficients for each pair to calculate the intensity of competition as the relative strength of interspecific and intraspecific competition: $\rho = \sqrt{\frac{\alpha_{ij}\alpha_{ji}}{\alpha_{ii}\alpha_{jj}}}$, where ρ can only include interaction coefficients with positive values (i.e. competition, not facilitation) and can range from 0 to infinity (also called 'niche overlap' following Chesson, 2013). If conspecifics limit themselves much more than they limit their competitors, values of ρ will be very low (i.e. close to 0). If conspecifics limit themselves and other species similarly, ρ will be high; values of $\rho < 1$ represent stronger stabilising effects on species coexistence (Adler et al., 2018; Letten et al., 2017). If the intensity of competition varies among species pairs, we hypothesize that communities that contain all species might diffuse competitive interactions (Barbier et al., 2021) providing a mechanism for positive biodiversity effects. Two main weaknesses of the Lotka–Volterra model are that it is not truly predictive (it can only be fitted to the observations a posteriori) and ignores functional and physiological aspects, therefore its predictions mostly concern density and biomass patterns rather than other functional processes, for example, energy production (Loreau, 2010).

2.5 | Statistical analyses on biomass production and energy fluxes

We used nonlinear growth models to describe changes in total biovolume (μm^3/μl) and net energy production (J/day) over time for each diversity treatment. First, growth models were fitted across all replicates within each diversity treatment to visualize qualitative differences in biomass and net energy production among monocultures, pairs and communities. We chose the best-fitting model among four candidates to allow for differences in the qualitative shapes of the time series, adopted from Malerba et al. (2018). The models were a logistic-type sinusoidal growth model with lower asymptote forced to 0 (i.e. three-parameter logistic curve), a logistic-type sinusoidal growth model with non-zero lower asymptote (i.e. four-parameter logistic curve), a Gompertz-type sinusoidal growth model (i.e. three-parameter Gompertz curve) and a modified Gompertz-type sinusoidal growth model including population decline after reaching a maximum (i.e. four-parameter Gompertz-like curve including mortality; see Table S2 for model structure). Akaike information criterion (AIC) were used to determine which growth model best described the dynamics of each diversity level. Successful convergence was ensured for all best-fitting models.

To formally test the effects of biodiversity on the maximum rates of increase ($r_{\max}$) and maximum values (K) of biovolume and net energy production we followed a three-step approach (Table S2; Malerba et al., 2018). First, we fitted the four growth models described above to each individual replicate culture and chose the best-fitting model among the four candidates to best describe changes in the total biovolume (μm^3/μl) or net energy production (J/day) of each culture over time. As above, we used AIC to determine which growth model best described the dynamics of a culture and successful convergence was ensured for all best-fitting models. Second, we used the best-fitting model to estimate growth parameters (i.e. $r_{\max}$ and K) of biovolume and net energy production for each culture. From each nonlinear curve, we extracted the maximum predicted value (*K*) of total biovolume ($\mu m^3/\mu l$) or net energy production (J/day). From the first derivative of the curve, we extracted the maximum rate of increase (r_{max}, unit: day^{-1}) in biovolume or net energy production. Because r_{max} and *K* are extracted from the shape of the model fit, and not from the model coefficients, these parameters are comparable even when estimated from models with different parametrizations (Malerba et al., 2018). Third, we used an analysis of covariance to evaluate the influence of species diversity on each parameter, using a linear model including the initial biovolume estimated from the previous step as a covariate and species diversity as a factor (three levels). Because r_{max} for biovolume and *K* for net energy had heterogeneous variances among diversity treatments, we used generalized least squares models (instead of linear models) including treatment-specific variance for each level of diversity (varIdent function in R). We then estimated and plotted least square means and 95% confidence intervals using Tukey *p*-value adjustment for comparing three estimates.

To estimate the biovolume dependence of energy fluxes, energy rates (J/hr per 5 ml sample) and total biovolume ($\mu m^3/\mu l$) were $\log_e$-transformed to fit a linear model including species diversity as predictor (factor with three levels), total biovolume as covariate and their interaction. We fit generalized-least squares models to allow for differences in variance among levels of diversity because variances were heterogeneous. We then used the model predictions for energy production (photosynthesis) and consumption (post-illumination and dark metabolism) to estimate net energy production over a 24-hr period (14 hr of energy produced through net photosynthesis minus 0.25 hr of energy consumed with post-illumination metabolism and 9.75 hr of dark metabolism). We also tested differences in the net daily energy production among species in monoculture as a function of their biovolume using a linear model with species and biovolume as predictors, including their interaction; both net energy (J/day) and biovolume ($\mu m^3/\mu l$) were $\log_e$-transformed. All analyses and plots were done in R version 3.5.0 using R Studio interface.

3 | RESULTS

3.1 | Successional patterns of biomass and net energy production

The production of biomass increased over time following a sinusoidal shape that was influenced by species diversity (Figure 1a). All diversity treatments approached carrying capacity towards the end of the experiment but at different rates. Communities showed faster rates of biovolume increase and a greater total biovolume. Changes in biovolume were best described by a three-parameter Gompertz model for monocultures and pairs, and by a Gompertz model with mortality for communities because their biovolume declined in the later part of the experiment (Table S3).

Net biodiversity effects confirmed that communities were on average more productive than monocultures and species pairs, and that the positive effect of diversity peaked mid-experiment (day 6) and decreased afterwards (Figure 1b). Both complementarity (Figure 1c) and selection effects (Figure 1d) enhanced biomass production in communities but species complementarity was the strongest driver. Selection effects progressively increased over time because the most productive species in monoculture dominated biovolume in communities (*Tetraselmis* sp., Figure S1). Despite this dominance, all species increased in abundance and none was competitively excluded (Figure S1). Species evenness declined from 1 to ~0.75 in both communities and pairs, albeit decline was up to 70% in some species pairs (Figure S1h). Net biodiversity effects in species pairs followed a similar pattern to that of communities but were only weakly positive (Figure 1b). For pairs, biodiversity effects were solely driven by complementarity (Figure 1c) as selection effects were on average zero (Figure 1d).

Net energy production also followed a sinusoidal shape but peaked earlier than biomass for all diversity treatments (~ day 3). Net energy production increased at faster rates in communities but the effects of diversity on energy were subtler than those on biomass (Figure 1e). Net energy production was best described by a three-parameter logistic curve for monocultures (zero lower asymptote) and four-parameter logistic curve for pairs and communities (non-zero lower asymptote; Table S3).

3.2 | Effects of biodiversity on rates of increase and maximum values of production

Species diversity affected both the maximum rate of increase (r_{max}) and maximum values (*K*) of biomass and net energy production but in different ways. Diversity enhanced the maximum rates of increase (r_{max}) in biovolume ($F_{2,46} = 5.83$, $p = 0.006$; Figure 2a) and net energy production ($F_{2,46} = 15.82$, $p < 0.0001$; Figure 2b) so that production rates augmented ~ twice as fast in communities than monocultures (biovolume: communities = 3.88×10^4 vs. monocultures = 2.38×10^4 μm^3 μl^{-1} day^{-1}, Table S4; net energy: communities = 5.45 vs. monocultures = 2.39 J/day, Table S5). Species pairs had intermediate rates of increase between monocultures and communities (Figure 2; Tables S4 and S5). Diversity also increased the maximum value of biomass: communities reached a greater total biovolume ($K = 18.8 \times 10^4$ $\mu m^3/\mu l$; $F_{2,46} = 3.64$, $p = 0.034$) than species pairs (14.6×10^4) and monocultures (14.5×10^4), which did not differ statistically (Figure 2c; Table S4). Species diversity, however, had no effect on maximum net energy production ($F_{2,46} = 0.68$, $p = 0.51$; Figure 2d; Table S5), which, however, tended to increase with diversity (monocultures = 8.43, pairs = 9.17, communities = 9.51 J/day). Differences in productivity and growth rates among species (see Section 3.4) meant that production rates were more variable among the 5 monocultures and 10 species pairs than the 5 communities, which contained the same pool of species (Figure 2a,c; Tables S6 and S7). The three replicates of each monoculture and pair, however,

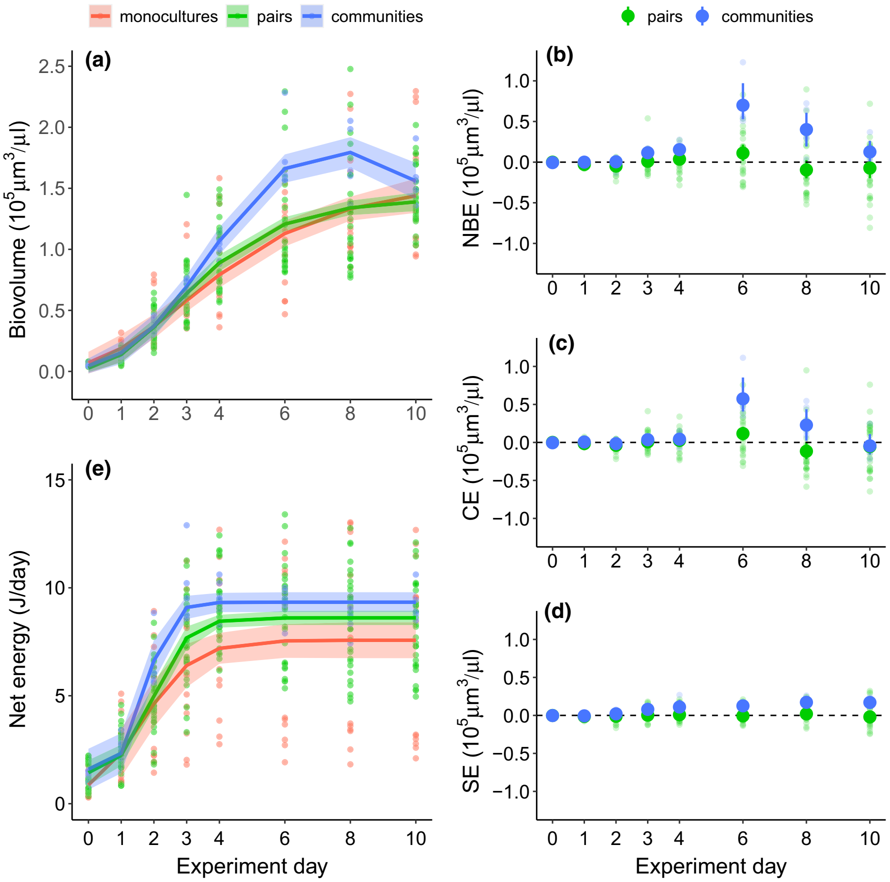

FIGURE 1 Temporal patterns of biovolume and net energy production at three levels of species diversity. (a) Communities reach a greater total biovolume than monocultures and species pairs ($N = 50$ for each experiment day). (b) Positive net biodiversity effects (NBEs) in communities result from both a positive complementarity effect (CE) that peaks mid-experiment (c), and a positive selection effect (SE) that increases over time (d). Net biodiversity effects in species pairs follow a similar pattern but are smaller in magnitude and only driven by complementarity effects. The point range (b, c and d) represents the population mean with 95% confidence limits ($N = 15$ on day 0 and $N = 35$ on other days). (e) Net energy production follows a logistic growth but peaks earlier than biovolume and, similarly, increases faster in more diverse mixtures ($N = 50$ for each experiment day, minus 1 data point lost for species pairs in day 2). Lines represent the fit value from the best-fitting model and 95% confidence interval (a, b). See Figure 2 for the formal comparison of maximum rates of increase (r_{max}) and maximum values (K) of biovolume and net energy production among diversity treatments

behaved consistently (Figure S1) and the confidence intervals for both biomass r and K were similar across levels of diversity (overall larger for communities because of their lower replication, Table S4).

3.3 | Biomass mediates the effects of biodiversity on energy fluxes

The effects of species diversity on energy fluxes were mediated by biovolume. At low biovolumes, diversity increased rates of energy production (photosynthesis; Figure 3a) and consumption (dark metabolism; Figure 3c). But this positive effect weakened as biovolume increased so that rates converged at larger biovolumes (i.e. communities had shallower slopes than monocultures; ln-biovolume × diversity interaction, Table S8). While the interaction between diversity and biovolume was significant for the energy consumed by post-illumination metabolism, post-hoc tests indicated that slopes were not statistically different and that communities had greater rates of post-illumination metabolism across the entire range of biovolume (Figure 3b). The energy fluxes measured for species pairs were intermediate and their slope was statistically different from that of monocultures and communities only for dark metabolism (Figure 3; Table S8). Finally, estimates of net daily energy production over a 24-hr period obtained from model predictions showed the overall same pattern: species diversity increased net energy production at low biovolumes but these effects weakened as biovolume accumulated (Figure 3d).

3.4 | Competitive interactions within and among species

The intensity of competition varied among species and generally increased with species size. Larger species, such as *Amphidinium* and *Tetraselmis*, suffered more from intraspecific than interspecific competition, and they had stronger competitive effects on smaller

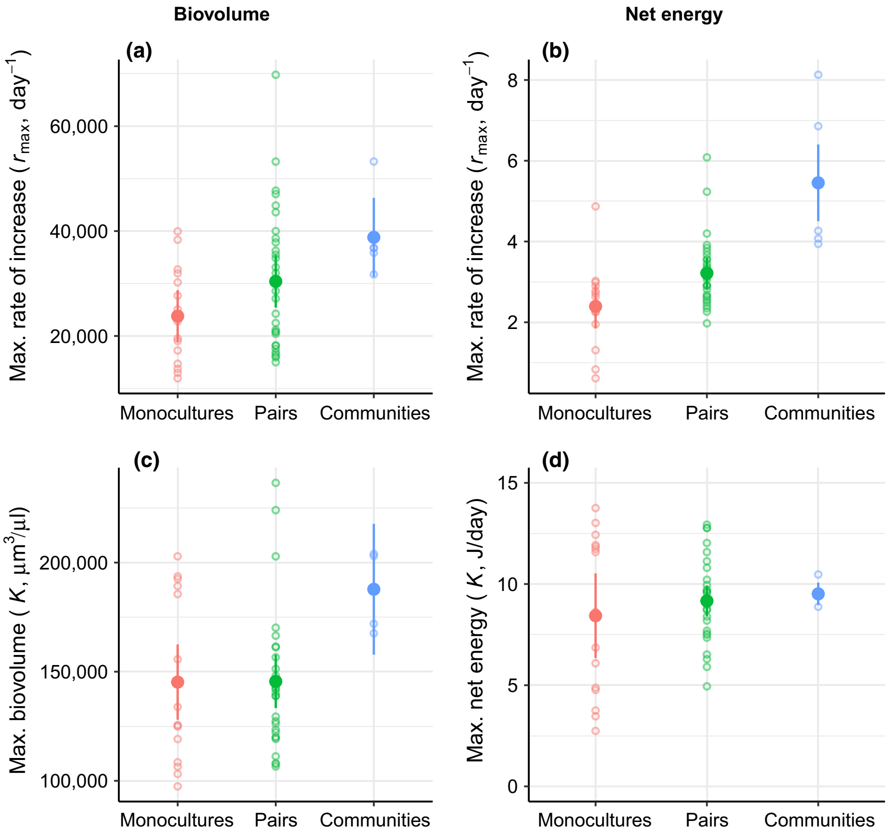

FIGURE 2 Mean growth parameters (r_{max} and K, +95% CI of the means, $N = 50$) of each diversity treatment (x-axis) for biovolume (left) and net energy production (right). The max. rate of increase of biovolume (a) or net energy production (b) indicates the maximum value of the first derivative of the best-fitting growth model (r_{max}). The max. rates of increase of biovolume and net energy production progressively increase with species diversity. Max. biovolume (c) or net energy (d) indicate the maximum Y-value in the best-fitting growth model (K). Communities reach greater max. biovolume than species pairs and monocultures, but there is no difference in the max. net energy produced. Empty points represent the growth parameters of each replicate, as estimated by fitting nonlinear growth models to each time series, after correcting for the effects of initial biovolume. Each point range represents the mean and 95% confidence interval from estimated marginal means

species (Figure 4a; Table S9; Figure S1 for size). The dinoflagellate *Amphidinium* had both the strongest intraspecific and interspecific competitive effects. The green alga *Tetraselmis* had the greatest intrinsic population growth rate (Figure 4b) and among the greatest rates of net energy production (second to *Dunaliella*, Figure 4c), explaining why it was the dominant species in mixtures and the most productive species overall in terms of biovolume. Net energy production increased with biovolume at different rates for each species (biovolume × species: $F_{4,110} = 5.47$, $p = 0.0005$); interestingly, energy fluxes showed stronger biomass dependence for larger species, such as *Tetraselmis* and *Amphidinium* (Figure 4c; Table S10). Competition intensity (measured as ρ, which represents the relative strength of interspecific and intraspecific competition) varied for each combination of species from a minimum of 0.15 to a maximum of 1.27 ($M \pm SE$: 0.81 ± 0.13). The dominant species *Tetraselmis* was the only one that strongly competed with all species (ρ close or above 1 for all species pairs; Figure 4d). We could not calculate ρ for the pair *Amphidinium*–*Dunaliella* because one of the competition coefficients was negative, indicating facilitation ($\alpha_{amp,dun}$).

4 | DISCUSSION

Our finding that the strength of biodiversity effects on biomass varies during community development aligns with studies in other systems (Armitage, 2016; Frank et al., 2020; Weis et al., 2007): positive BEF effects initially increase, reaching an intermediate peak, and then decline in older communities. We extend these findings by identifying the coincident trajectories of community energy fluxes—importantly, these fluxes are not perfectly coupled to biomass effects. Diverse mixtures produce net energy at faster rates, producing biomass faster and accumulating more total biomass but with no difference in maximum energy production (Figure 5). The idea that diversity enhances biomass production by increasing the flux of energy is intuitive but actual examples are rare (Barnes et al., 2020), and none have explored how biodiversity changes these fluxes over time. Our results show that the effects of diversity on energy fluxes are temporally dynamic and saturate as biomass accumulates. So, while the efficiency of biomass production increases with diversity, as long as resource partitioning is possible (Hodapp et al., 2019), energy production seems subject to stronger constraints.

The saturating pattern of biodiversity effects and the initial higher rates of energy use in mixtures indicate that biodiversity can enhance functioning through a dilution of intraspecific competition, which is a consequence of the substitutive experimental design (50% and 80% less conspecifics in pairs and communities respectively). Metabolic suppression is stronger in response to conspecifics than heterospecifics (Armitage, 2016; Ghedini, Malerba, et al., 2020), similarly to competition for resources (Adler et al., 2018; Weis et al., 2007; Weisser et al., 2017) as we also observe for most species (Figure 4d). Since metabolic suppression occurs even when resources are not limiting (Lovass et al., 2020),

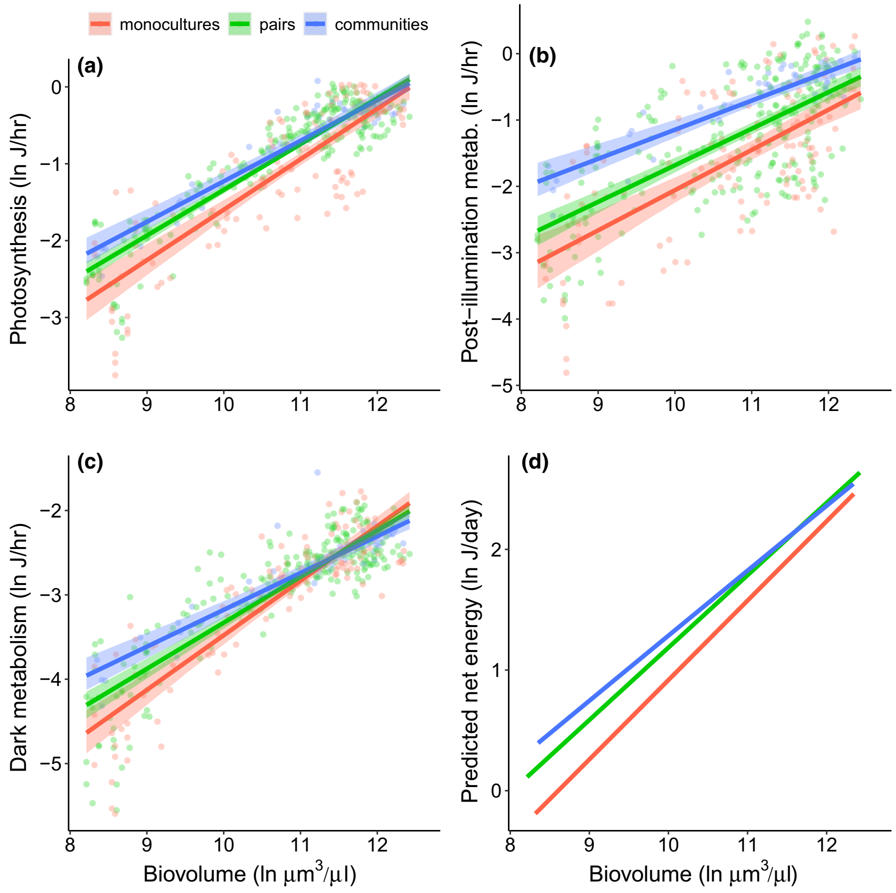

FIGURE 3 Total biovolume mediates the effects of biodiversity on energy fluxes. The energy produced through (a) net photosynthesis and consumed through (b) post-illumination and (c) dark metabolism increases with total biovolume but at different rates across diversity treatments (ln-transformed rates shown for 5 ml vials, $N = 50$ at each of eight sampling days for each rate, minus one sample lost for photosynthesis for day 2). While species diversity increases energy fluxes at low biovolumes, these effects weaken as biovolume increases. Lines represent the best-fitting value and 95% confidence interval from generalized least-squares models on $\log_e$-transformed data, allowing for differences in variance among diversity levels. Note that slopes are not different for post-illumination metabolism (b; Table S8). (d) Net daily energy production estimated from photosynthetic and metabolic rates over a 24-hr period follows the same pattern (calculated on a 14–10 hr light-dark cycle)

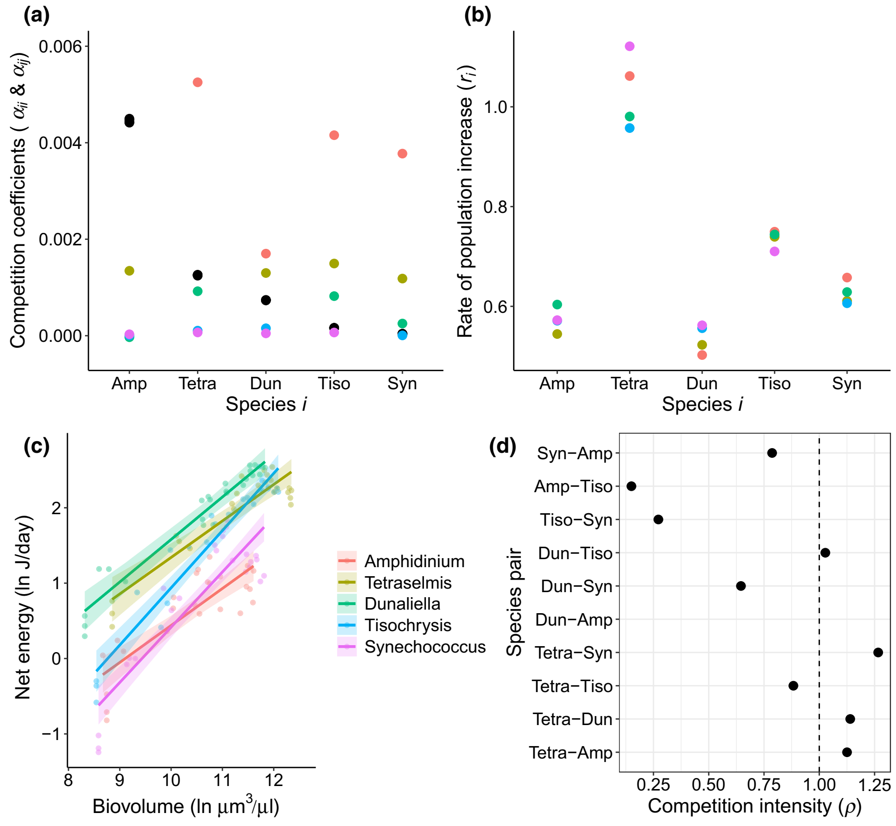

FIGURE 4 Differences in species competitive ability. (a) Comparison of intraspecific and interspecific competition coefficients for each species pair calculated from Lotka–Volterra models using monoculture and biculture data on cell abundance (cells μl^{-1} day^{-1}; Table S9). Black points represent the intraspecific competition coefficient α_{ii} for each species *i* (*x*-axis, decreasing size), while coloured points represent the effect of species *j* (see legend for colour) on species *i* (i.e. α_{ij}). (b) Species intrinsic rate of increase r_i estimated from competition models and (c) rates of daily net energy production as a function of species biovolume in monoculture. (d) Competition intensity ('niche overlap' following Chesson, 2013) quantifying the relative strength of interspecific and intraspecific competition for each combination of two species ($\rho = \sqrt{\frac{\alpha_{ij}\alpha_{ji}}{\alpha_{ii}\alpha_{jj}}}$)

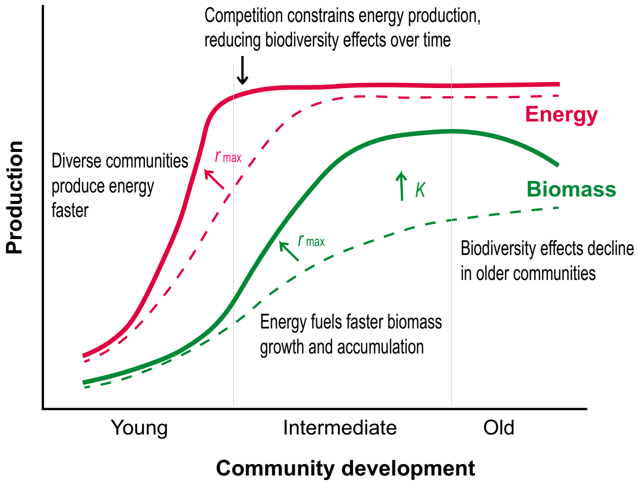

FIGURE 5 Conceptual figure showing how more diverse communities (solid lines) produce net energy (magenta) and biomass (green) at faster rates (r_{max}) than low-diversity communities (dashed lines), thus allowing greater biomass accumualtion (*K*). As biomass accumulates and competition intesifies, however, metabolic density dependence reduces energy and thus biomass production so that the positive effects of biodiversity progressively decline in older communities

mixtures initially produce net energy and biomass faster because each species competes with fewer conspecifics. But as biomass accumulates, communities suffer stronger metabolic suppression (shallower slopes of energy flux with biovolume) which reduces production.

Why are energy fluxes under stronger biomass dependence in communities? One obvious reason is the increase in the dominance of one (or few) species over time (Figure S1f; Ghedini et al., 2018). The species that dominated communities, *Tetraselmis*, had faster growth rates and higher net energy production than most other species, but also a stronger biomass dependence of production. Its progressive dominance therefore impacted competitors and constrained productivity as indicated by the increase in selection effects and reduction in complementarity effects over time, which is expected in stable environments (Hodapp et al., 2016). Natural phytoplankton assemblages show similar positive selection effects where the identity and traits of dominant species affect BEF relationships (Hodapp et al., 2015; Otero et al., 2020). Nonetheless, biodiversity effects were up to five times stronger in communities than pairs. Even a small increase in diversity (two to five species) can alleviate the cost of competition by allowing strong competitors to partition their impacts on other species (Barbier et al., 2020; Maynard et al., 2017). Partitioning is possible in our system because species spanned diverse groups and sizes correlated with differences in nutrient uptake, storage and photosynthetic efficiency (Litchman et al., 2007; Malerba et al., 2018) which resulted in diverse competitive interactions.

Another mechanism could explain the steeper decline of energy fluxes with biomass in communities: if heterospecific and intraspecific effects on energy use converge over time. Convergence may occur because conspecifics diversify their resource use to reduce intraspecific competition (Svanbäck & Bolnick, 2007) or because species converge on similar resource-use strategies (Macarthur & Levins, 1967; Mason et al., 2012; TerHorst et al., 2010). While we can only speculate on this mechanism (because we do not have access to species-specific metabolic rates in mixtures), trait convergence could reduce fitness differences (Narwani et al., 2017) and explain why older assemblages often support functionally similar species (Leopold & Fukami, 2020; Mason et al., 2012). Coadaptation might thus facilitate coexistence but weaken diversity effects, explaining some variation in the strength and direction of BEF relationships among studies (Aubree et al., 2020; Chen et al., 2019). For instance, artificially assembled communities might contain species with greater trait diversity than natural communities that have been exposed to longer environmental filtering (Gerhard et al., 2021). Since our experiment only covered ~10 generations, evolutionary changes probably played a minor role. But we cannot preclude phenotypic changes in metabolism because resource use and metabolic rates are plastic (Lovass et al., 2020; Poulson-Ellestad et al., 2014; Svanbäck & Bolnick, 2007).

The benefits of biodiversity on production declined rapidly as cultures approached carrying capacity (within 10 days). Similarly, Frank et al. (2020) found that mixtures of freshwater phytoplankton produced more biomass than monocultures during exponential growth but underyielded at carrying capacity. A decline in production is typical of mature systems but is usually observed on decadal time-scales in nature (Boit & Gaedke, 2014; Lasky et al., 2014; Mason et al., 2012) because natural systems are continuously disturbed by fluctuations in nutrient availability, grazers and species immigration that can alter BEF relationships (Smith et al., 2016; Vallina et al., 2014). Our simplified communities, instead, developed in a stable and undisturbed environment where competition (particularly for light) increased rapidly due to biomass growth and self-shading (Malerba et al., 2018); hence, our communities might approximate the characteristics of some systems more than others. Despite this simplicity, the patterns of biomass and net energy production we observe align with succession theory (Odum, 1969) and the successional effects of biodiversity are consistent with those reported for phytoplankton (Weis et al., 2007), forest communities (Lasky et al., 2014) and bacteria (Armitage, 2016). The reproducibility of BEF relationships across diverse systems indicates that the temporal effects of biodiversity we observe may approximate those experienced by natural communities during succession.

5 | CONCLUSIONS

Biodiversity affects net energy and biomass production differently during community development. In younger assemblages, diversity increases the rates at which net energy and, thus, biomass grow by diffusing competitive effects. As biomass accumulates and stronger competitors dominate, energy fluxes decline at faster rates in mixtures so that biodiversity effects weaken over time. If biodiversity effects are stronger in younger communities, then the periodic removal of species typical of long-term field experiments (Balvanera et al., 2006) may partly explain why BEF relationships strengthen over time in these systems (i.e. by interrupting succession; Cardinale et al., 2007; Fargione et al., 2007; Meyer et al., 2016). Other factors, however, may contribute to the dramatic increase in positive BEF relationships that are not captured in our experiment. First, complementarity in resource use might strengthen over longer-time scales, enhancing efficiency and productivity—critically, these effects seem stronger in mixtures than monocultures (Lawrence et al., 2012). Second, increased complementarity can result in ecosystem feedbacks that support productivity over time, particularly in nutrient-poor systems. For instance, plant diversity can increase nitrogen cycling and availability in the soil boosting diversity–productivity relationships (Dybzinski et al., 2008; Reich et al., 2012). These ecosystem feedbacks might be particularly important for relatively closed, undisturbed systems (such as many terrestrial communities) and less so for highly dynamic systems, such as oceanic phytoplankton communities which are often mixed by water currents. Hence, a variety of mechanisms may play out to alter the strength and direction of BEF relationships over time, and their importance might differ among systems. As our findings suggest, energy fluxes can provide a general underlying mechanism to interpret BEF relationships and compare the effects of biodiversity across systems at different stages of development and disturbance regimes.

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AUTHORS' CONTRIBUTIONS

G.G. and M.L. designed the study; G.G. conducted the experiment and analysed the data with feedback from D.J.M.; G.G. drafted the manuscript. All authors contributed to revisions.

COMPETING OF INTEREST

G.G. and D.J.M. are Associate Editors of Functional Ecology, but took no part in the peer review and decision-making processes for this paper. We do not have any other competing interest.

DATA AVAILABILITY STATEMENT

Data available on Figshare https://doi.org/10.26180/16665964 (Ghedini et al., 2021).

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REFERENCES

Adler, P. B., Smull, D., Beard, K. H., Choi, R. T., Furniss, T., Kulmatiski, A., Meiners, J. M., Tredennick, A. T., & Veblen, K. E. (2018). Competition and coexistence in plant communities: Intraspecific competition is stronger than interspecific competition. *Ecology Letters*, *21*, 1319–1329. https://doi.org/10.1111/ele.13098

Armitage, D. W. (2016). Time-variant species pools shape competitive dynamics and biodiversity-ecosystem function relationships. *Proceedings of the Royal Society B: Biological Sciences*, *283*, 20161437. https://doi.org/10.1098/rspb.2016.1437

Aubree, F., David, P., Jarne, P., Loreau, M., Mouquet, N., & Calcagno, V. (2020). How community adaptation affects biodiversity–ecosystem functioning relationships. *Ecology Letters*, *23*, 1263–1275. https://doi.org/10.1111/ele.13530

Balvanera, P., Pfisterer, A. B., Buchmann, N., He, J. S., Nakashizuka, T., Raffaelli, D., & Schmid, B. (2006). Quantifying the evidence for biodiversity effects on ecosystem functioning and services. *Ecology Letters*, *9*, 1146–1156. https://doi.org/10.1111/j.1461-0248.2006.00963.x

Barbier, M., de Mazancourt, C., Loreau, M., & Bunin, G. (2020). Fingerprints of high-dimensional coexistence in complex ecosystems. *bioRxiv*, 652230.

Barbier, M., de Mazancourt, C., Loreau, M., & Bunin, G. (2021). Fingerprints of high-dimensional coexistence in complex ecosystems. *Physical Review X*, *11*, 011009. https://doi.org/10.1103/PhysRevX.11.011009

Barnes, A. D., Jochum, M., Lefcheck, J. S., Eisenhauer, N., Scherber, C., O'Connor, M. I., de Ruiter, P., & Brose, U. (2018). Energy flux: The link between multitrophic biodiversity and ecosystem functioning. *Trends in Ecology & Evolution*, *33*, 186–197. https://doi.org/10.1016/j.tree.2017.12.007

Barnes, A. D., Scherber, C., Brose, U., Borer, E. T., Ebeling, A., Gauzens, B., Giling, D. P., Hines, J., Isbell, F., Ristok, C., Tilman, D., Weisser, W. W., & Eisenhauer, N. (2020). Biodiversity enhances the multitrophic control of arthropod herbivory. *Science Advances*, *6*, eabb6603. https://doi.org/10.1126/sciadv.abb6603

Beardall, J., Burger-Wiersma, T., Rijkeboer, M., Sukenik, A., Lemoalle, J., Dubinsky, Z., & Fontvielle, D. (1994). Studies on enhanced post-illumination respiration in microalgae. *Journal of Plankton Research*, *16*, 1401–1410. https://doi.org/10.1093/plankt/16.10.1401

Boit, A., & Gaedke, U. (2014). Benchmarking successional progress in a quantitative food web. *PLoS ONE*, *9*, e90404. https://doi.org/10.1371/journal.pone.0090404

Brose, U., & Hillebrand, H. (2016). Biodiversity and ecosystem functioning in dynamic landscapes. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *371*, 20150267. https://doi.org/10.1098/rstb.2015.0267

Burson, A., Stomp, M., Greenwell, E., Grosse, J., & Huisman, J. (2018). Competition for nutrients and light: Testing advances in resource competition with a natural phytoplankton community. *Ecology*, *99*, 1108–1118. https://doi.org/10.1002/ecy.2187

Buzhdygan, O. Y., Meyer, S. T., Weisser, W. W., Eisenhauer, N., Ebeling, A., Borrett, S. R., Buchmann, N., Cortois, R., De Deyn, G. B., de Kroon, H., Gleixner, G., Hertzog, L. R., Hines, J., Lange, M., Mommer, L., Ravenek, J., Scherber, C., Scherer-Lorenzen, M., Scheu, S., … Petermann, J. S. (2020). Biodiversity increases multitrophic energy use efficiency, flow and storage in grasslands. *Nature Ecology & Evolution*, *4*, 393–405. https://doi.org/10.1038/s41559-020-1123-8

Cardinale, B. J., Duffy, J. E., Gonzalez, A., Hooper, D. U., Perrings, C., Venail, P., Narwani, A., Mace, G. M., Tilman, D., Wardle, D. A., Kinzig, A. P., Daily, G. C., Loreau, M., Grace, J. B., Larigauderie, A., Srivastava, D. S., & Naeem, S. (2012). Biodiversity loss and its impact on humanity. *Nature*, *486*, 59. https://doi.org/10.1038/nature11148

Cardinale, B. J., Wright, J. P., Cadotte, M. W., Carroll, I. T., Hector, A., Srivastava, D. S., Loreau, M., & Weis, J. J. (2007). Impacts of plant diversity on biomass production increase through time because of species complementarity. *Proceedings of the National Academy of Sciences of the United States of America*, *104*, 18123–18128. https://doi.org/10.1073/pnas.0709069104

Chen, B., Smith, S. L., & Wirtz, K. W. (2019). Effect of phytoplankton size diversity on primary productivity in the North Pacific: Trait distributions under environmental variability. *Ecology Letters*, *22*, 56–66. https://doi.org/10.1111/ele.13167

Chesson, P. (2013). Species competition and predation. In R. Leemans (Ed.), *Ecological systems* (pp. 223–256). Springer.

Dybzinski, R., Fargione, J. E., Zak, D. R., Fornara, D., & Tilman, D. (2008). Soil fertility increases with plant species diversity in a long-term biodiversity experiment. *Oecologia*, *158*, 85–93. https://doi.org/10.1007/s00442-008-1123-x

Fargione, J., Tilman, D., Dybzinski, R., Lambers, J. H. R., Clark, C., Harpole, W. S., Knops, J. M. H., Reich, P. B., & Loreau, M. (2007). From selection to complementarity: Shifts in the causes of biodiversity-productivity relationships in a long-term biodiversity experiment. *Proceedings of the Royal Society B: Biological Sciences*, *274*, 871–876.

Frank, F., Danger, M., Hillebrand, H., & Striebel, M. (2020). Stoichiometric constraints on phytoplankton resource use efficiency in monocultures and mixtures. *Limnology and Oceanography*, *65*, 1734–1746. https://doi.org/10.1002/lno.11415

Gatto, M. (1990). A general minimum principle for competing populations: Some ecological and evolutionary consequences. *Theoretical Population Biology*, *37*, 369–388. https://doi.org/10.1016/0040-5809(90)90044-V

Gerhard, M., Mori, C., & Striebel, M. (2021). Nonrandom species loss in phytoplankton communities and its effect on ecosystem functioning. *Limnology and Oceanography*, *66*, 779–792. https://doi.org/10.1002/lno.11642

Ghedini, G., Loreau, M., & Marshall, D. J. (2020). Community efficiency during succession: A test of MacArthur's minimisation principle in phytoplankton communities. *Ecology*, *101*, e03015.

Ghedini, G., Malerba, M. E., & Marshall, D. J. (2020). How to estimate community energy flux? A comparison of approaches reveals that size-abundance trade-offs alter the scaling of community energy flux. *Proceeding of the Royal Society Biological Sciences*, *287*, 20200995. https://doi.org/10.1098/rspb.2020.0995

Ghedini, G., Marshall, D. J., & Loreau, M. (2021). Data from: Phytoplankton diversity affects biomass and energy production differently during community development. *Figshare*, https://doi.org/10.26180/16665964

Ghedini, G., White, C. R., & Marshall, D. J. (2017). Does energy flux predict density-dependence? An empirical field test. *Ecology*, *98*, 3116–3126. https://doi.org/10.1002/ecy.2033

Ghedini, G., White, C. R., & Marshall, D. J. (2018). Metabolic scaling across succession: Do individual rates predict community-level energy use? *Functional Ecology*, *32*, 1447–1456. https://doi.org/10.1111/1365-2435.13103

Gonzalez, A., Germain, R. M., Srivastava, D. S., Filotas, E., Dee, L. E., Gravel, D., Thompson, P. L., Isbell, F., Wang, S., Kéfi, S., Montoya, J., Zelnik, Y. R., & Loreau, M. (2020). Scaling-up biodiversity-ecosystem functioning research. *Ecology Letters*, *23*, 757–776. https://doi.org/10.1111/ele.13456

Guillard, R. R. L., & Ryther, J. H. (1962). Studies of marine planktonic diatoms. I. *Cyclotella nana* Hustedt and *Detonula confervacea* Cleve. *Canadian Journal of Microbiology*, *8*, 229–239.

Hillebrand, H., Dürselen, C. D., Kirschtel, D., Pollingher, U., & Zohary, T. (1999). Biovolume calculation for pelagic and benthic microalgae. *Journal of Phycology*, *35*, 403–424. https://doi.org/10.1046/j.1529-8817.1999.3520403.x

Hodapp, D., Hillebrand, H., Blasius, B., & Ryabov, A. B. (2016). Environmental and trait variability constrain community structure and the biodiversity-productivity relationship. *Ecology*, *97*, 1463–1474. https://doi.org/10.1890/15-0730.1

Hodapp, D., Hillebrand, H., & Striebel, M. (2019). 'Unifying' the concept of resource use efficiency in ecology. *Frontiers in Ecology and Evolution*, *6*, 233. https://doi.org/10.3389/fevo.2018.00233

Hodapp, D., Meier, S., Muijsers, F., Badewien, T. H., & Hillebrand, H. (2015). Structural equation modeling approach to the diversity-productivity relationship of Wadden Sea phytoplankton. *Marine Ecology Progress Series*, *523*, 31–40. https://doi.org/10.3354/meps11153

Hooper, D. U., Chapin, F. S., Ewel, J. J., Hector, A., Inchausti, P., Lavorel, S., Lawton, J. H., Lodge, D. M., Loreau, M., Naeem, S., Schmid, B., Setala, H., Symstad, A. J., Vandermeer, J., & Wardle, D. A. (2005). Effects of biodiversity on ecosystem functioning: A consensus of current knowledge. *Ecological Monographs*, *75*, 3–35. https://doi.org/10.1890/04-0922

Inouye, B. D. (2001). Response surface experimental designs for investigating interspecific competition. *Ecology*, *82*, 2696–2706. https://doi.org/10.1890/0012-9658(2001)082[2696:RSEDFI]2.0.CO;2

Isbell, F., Craven, D., Connolly, J., Loreau, M., Schmid, B., Beierkuhnlein, C., Bezemer, T. M., Bonin, C., Bruelheide, H., de Luca, E., Ebeling, A., Griffin, J. N., Guo, Q., Hautier, Y., Hector, A., Jentsch, A., Kreyling, J., Lanta, V., Manning, P., … Eisenhauer, N. (2015). Biodiversity increases the resistance of ecosystem productivity to climate extremes. *Nature*, *526*, 574–577. https://doi.org/10.1038/nature15374

Jeffrey, S. W., & LeRoi, J. M. (1997). Simple procedures for growing SCOR reference microalgal cultures. In S. W. Jeffrey, R. F. C. Mantoura, & S. W. Wright (Eds.), *Phytoplankton pigments in oceanography; Monographs on oceanographic methodology* (pp. 181–205). UNESCO.

Lasky, J. R., Uriarte, M., Boukili, V. K., Erickson, D. L., John Kress, W., & Chazdon, R. L. (2014). The relationship between tree biodiversity and biomass dynamics changes with tropical forest succession. *Ecology Letters*, *17*, 1158–1167. https://doi.org/10.1111/ele.12322

Lawrence, D., Fiegna, F., Behrends, V., Bundy, J. G., Phillimore, A. B., Bell, T., & Barraclough, T. G. (2012). Species interactions alter evolutionary responses to a novel environment. *PLOS Biology*, *10*, e1001330. https://doi.org/10.1371/journal.pbio.1001330

Laws, E. A. (2013). Evaluation of in situ phytoplankton growth rates: A synthesis of data from varied approaches. *Annual Review of Marine Science*, *5*, 247–268. https://doi.org/10.1146/annurev-marine-121211-172258

Leopold, D. R., & Fukami, T. (2020). Greater local diversity under older species pools may arise from enhanced competitive equivalence. *Ecology Letters*, *24*, 310–318.

Letten, A. D., Ke, P.-J., & Fukami, T. (2017). Linking modern coexistence theory and contemporary niche theory. *Ecological Monographs*, *87*, 161–177. https://doi.org/10.1002/ecm.1242

Lindeman, R. L. (1942). The trophic-dynamic aspect of ecology. *Ecology*, *23*, 399–418. https://doi.org/10.2307/1930126

Litchman, E., Klausmeier, C. A., Schofield, O. M., & Falkowski, P. G. (2007). The role of functional traits and trade-offs in structuring phytoplankton communities: Scaling from cellular to ecosystem level. *Ecology Letters*, *10*, 1170–1181. https://doi.org/10.1111/j.1461-0248.2007.01117.x

Loreau, M. (2010). *From populations to ecosystems: Theoretical foundations for a new ecological synthesis*. Princeton University Press.

Loreau, M., & Hector, A. (2001). Partitioning selection and complementarity in biodiversity experiments. *Nature*, *412*, 72–76. https://doi.org/10.1038/35083573

Loreau, M., Sapijanskas, J., Isbell, F., & Hector, A. (2012). Niche and fitness differences relate the maintenance of diversity to ecosystem function: Comment. *Ecology*, *93*, 1482–1487.

Lovass, M., Marshall, D. J., & Ghedini, G. (2020). Conspecific chemical cues drive density-dependent metabolic suppression independently of resource intake. *Journal of Experimental Biology*, *223*, jeb.224824. https://doi.org/10.1242/jeb.224824

MacArthur, R. (1969). Species packing, and what competition minimizes. *Proceedings of the National Academy of Sciences of the United States of America*, *64*, 1369–1371. https://doi.org/10.1073/pnas.64.4.1369

Macarthur, R., & Levins, R. (1967). The limiting similarity, convergence, and divergence of coexisting species. *The American Naturalist*, *101*, 377–385. https://doi.org/10.1086/282505

Malerba, M. E., Palacios, M. M., & Marshall, D. J. (2018). Do larger individuals cope with resource fluctuations better? An artificial selection approach. *Proceedings of the Royal Society B: Biological Sciences*, *285*(1884), 20181347. https://doi.org/10.1098/rspb.2018.1347

Malerba, M. E., Palacios, M. M., Palacios Delgado, Y. M., Beardall, J., & Marshall, D. J. (2018). Cell size, photosynthesis and the package effect: An artificial selection approach. *New Phytologist*, *219*, 449–461. https://doi.org/10.1111/nph.15163

Malerba, M. E., White, C. R., & Marshall, D. J. (2017). Phytoplankton size-scaling of net-energy flux across light and biomass gradients. *Ecology*, *98*, 3106–3115. https://doi.org/10.1002/ecy.2032

Mason, N. W. H., Richardson, S. J., Peltzer, D. A., de Bello, F., Wardle, D. A., & Allen, R. B. (2012). Changes in coexistence mechanisms along a long-term soil chronosequence revealed by functional trait diversity. *Journal of Ecology*, *100*, 678–689. https://doi.org/10.1111/j.1365-2745.2012.01965.x

Maynard, D. S., Crowther, T. W., & Bradford, M. A. (2017). Competitive network determines the direction of the diversity–function relationship. *Proceedings of the National Academy of Sciences of the United States of America*, *114*, 11464–11469. https://doi.org/10.1073/pnas.1712211114

Meyer, S. T., Ebeling, A., Eisenhauer, N., Hertzog, L., Hillebrand, H., Milcu, A., Pompe, S., Abbas, M., Bessler, H., Buchmann, N., De Luca, E., Engels, C., Fischer, M., Gleixner, G., Hudewenz, A., Klein, A.-M., de Kroon, H., Leimer, S., Loranger, H., … Weisser, W. W. (2016). Effects of biodiversity strengthen over time as ecosystem functioning declines at low and increases at high biodiversity. *Ecosphere*, *7*, e01619. https://doi.org/10.1002/ecs2.1619

Narwani, A., Bentlage, B., Alexandrou, M. A., Fritschie, K. J., Delwiche, C., Oakley, T. H., & Cardinale, B. J. (2017). Ecological interactions and coexistence are predicted by gene expression similarity in freshwater green algae. *Journal of Ecology*, *105*, 580–591. https://doi.org/10.1111/1365-2745.12759

Odum, E. P. (1969). Strategy of ecosystem development. *Science*, *164*, 262–270.

Otero, J., Álvarez-Salgado, X. A., & Bode, A. (2020). Phytoplankton diversity effect on ecosystem functioning in a coastal upwelling system. *Frontiers in Marine Science*, *7*, 592255. https://doi.org/10.3389/fmars.2020.592255

Poulson-Ellestad, K. L., Jones, C. M., Roy, J., Viant, M. R., Fernández, F. M., Kubanek, J., & Nunn, B. L. (2014). Metabolomics and proteomics reveal impacts of chemically mediated competition on marine plankton. *Proceedings of the National Academy of Sciences of the United States of America*, *111*, 9009–9014. https://doi.org/10.1073/pnas.1402130111

Reich, P. B., Tilman, D., Isbell, F., Mueller, K., Hobbie, S. E., Flynn, D. F. B., & Eisenhauer, N. (2012). Impacts of biodiversity loss escalate through time as redundancy fades. *Science*, *336*, 589–592. https://doi.org/10.1126/science.1217909

Schindelin, J., Arganda-Carreras, I., Frise, E., Kaynig, V., Longair, M., Pietzsch, T., Preibisch, S., Rueden, C., Saalfeld, S., Schmid, B., Tinevez, J.-Y., White, D. J., Hartenstein, V., Eliceiri, K., Tomancak, P., & Cardona, A. (2012). Fiji: An open-source platform for biological-image analysis. *Nature Methods*, *9*, 676–682. https://doi.org/10.1038/nmeth.2019

Six, C., Thomas, J. C., Brahamsha, B., Lemoine, Y., & Partensky, F. (2004). Photophysiology of the marine cyanobacterium *Synechococcus* sp. WH8102, a new model organism. *Aquatic Microbial Ecology*, *35*, 17–29. https://doi.org/10.3354/ame035017

Smith, S. L., Vallina, S. M., & Merico, A. (2016). Phytoplankton size-diversity mediates an emergent trade-off in ecosystem functioning for rare versus frequent disturbances. *Scientific Reports*, *6*, 34170. https://doi.org/10.1038/srep34170

Svanbäck, R., & Bolnick, D. I. (2007). Intraspecific competition drives increased resource use diversity within a natural population. *Proceedings of the Royal Society of London B: Biological Sciences*, *274*, 839–844. https://doi.org/10.1098/rspb.2006.0198

TerHorst, C. P., Miller, T. E., & Powell, E. (2010). When can competition for resources lead to ecological equivalence? *Evolutionary Ecology Research*, *12*, 843–854.

Tilman, D., Knops, J., Wedin, D., Reich, P., Ritchie, M., & Siemann, E. (1997). The influence of functional diversity and composition on ecosystem processes. *Science*, *277*, 1300. https://doi.org/10.1126/science.277.5330.1300

Vallina, S. M., Follows, M. J., Dutkiewicz, S., Montoya, J. M., Cermeno, P., & Loreau, M. (2014). Global relationship between phytoplankton diversity and productivity in the ocean. *Nature Communications*, *5*, 4299. https://doi.org/10.1038/ncomms5299

Wacker, L., Baudois, O., Eichenberger-Glinz, S., & Schmid, B. (2009). Diversity effects in early- and mid-successional species pools along a nitrogen gradient. *Ecology*, *90*, 637–648. https://doi.org/10.1890/07-1946.1

Weis, J. J., Cardinale, B. J., Forshay, K. J., & Ives, A. R. (2007). Effects of species diversity on community biomass production change over the course of succession. *Ecology*, *88*, 929–939. https://doi.org/10.1890/06-0943

Weisser, W. W., Roscher, C., Meyer, S. T., Ebeling, A., Luo, G., Allan, E., Beßler, H., Barnard, R. L., Buchmann, N., Buscot, F., Engels, C., Fischer, C., Fischer, M., Gessler, A., Gleixner, G., Halle, S., Hildebrandt, A., Hillebrand, H., de Kroon, H., … Eisenhauer, N. (2017). Biodiversity effects on ecosystem functioning in a 15-year grassland experiment: Patterns, mechanisms, and open questions. *Basic and Applied Ecology*, *23*, 1–73. https://doi.org/10.1016/j.baae.2017.06.002

White, C. R., Kearney, M. R., Matthews, P. G. D., Kooijman, S. A. L. M., & Marshall, D. J. (2011). A manipulative test of competing theories for metabolic scaling. *American Naturalist*, *178*, 746–754. https://doi.org/10.1086/662666

Williams, P. J. L. B., & Laurens, L. M. L. (2010). Microalgae as biodiesel & biomass feedstocks: Review & analysis of the biochemistry, energetics & economics. *Energy & Environmental Science*, *3*, 554–590.

Yang, X., Liu, L., Yin, Z., Wang, X., Wang, S., & Ye, Z. (2020). Quantifying photosynthetic performance of phytoplankton based on photosynthesis–irradiance response models. *Environmental Sciences Europe*, *32*, 24. https://doi.org/10.1186/s12302-020-00306-9

SUPPORTING INFORMATION

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