**Letters**

**Mismatches in thermal and nutrient physiology predict competitive outcomes among phytoplankton**

**Short running title**: Physiological mismatches predict competition

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# Abstract

Variations in temperature affect species fitness both directly, through changes in their physiology, as well as indirectly, by impacting their interaction with other species. To better predict the consequences of future climate change, it is thus crucial to understand how changes in temperature affect species interactions. Recent theoretical studies have demonstrated the potential for mismatches between prey and predators’ thermal physiology to alter consumer-resource dynamics. However, less attention has been paid to explaining interspecific competition, and, to our knowledge, no large experimental study has attempted to link theory and experiments. Here we investigated how mismatches in the thermal physiology of competing species affected the outcome of the competition in phytoplankton. We developed a theoretical model to investigate competition between species, and tested the predictions of this model against a large scale competition experiment of six species of freshwater phytoplankton at two temperatures and two nutrient conditions. We show that for five out of six species, competitive outcomes are well predicted by the theory based on mismatches of individual species’ traits. Furthermore, competitive outcome was sometimes dependent on temperature; these reversals were also well captured by the theory.

# Introduction

Climate change is predicted to be a major cause of species extinctions over the next century (Field *et al.* 2014), and a considerable threat to biodiversity (Thomas *et al.* 2004; Bellard *et al.* 2012). Susceptibility to climate change will depend on species’ environmental tolerances (Pacifici *et al.* 2015), with those occupying narrower thermal niches expected to be more vulnerable to climate warming (Magozzi & Calosi 2015). However, recent studies have highlighted that species interactions may play a greater role in mediating the impacts of climate change on populations than physiological tolerance limits (Dunn *et al.* 2009; Bellard *et al.* 2012; Cahill *et al.* 2013; Field *et al.* 2014). Indeed the key drivers of global change (warming, CO2 and nutrient enrichment) are known to affect various types of species interactions, including competition (Tylianakis *et al.* 2008). To better predict the consequences of future climate change, it is therefore crucial to understand how increased temperatures affect species interactions (Bestion & Cote 2017).

Metabolism sets the pace of life (Brown *et al.* 2004) and dictates a host of life-history traits and attributes that determine fitness, including population growth rate (Savage *et al.* 2004), abundance, mortality and interspecific interactions (Dell *et al.* 2011). Species vary widely in the way in which their metabolism and associated traits respond to temperature (Kingsolver 2009; Dell *et al.* 2011), and these differences in thermal physiology can greatly impact species interactions (Reuman *et al.* 2014; Dell *et al.* 2014). Mismatches can arise when species’ metabolic traits differ in their magnitude (the elevation of the thermal performance curve), relative rate of increase in metabolism with temperature (the slope of their temperature-ln performance relationship) and/or thermal optima (the temperature at which the performance is maximised) (Kordas *et al.* 2011). Recent theory suggests that mismatches in the thermal responses of body velocity between interacting species can play a key role in shaping the effects of temperature on consumer-resource dynamics (Dell *et al.* 2014). Mismatches in the temperature-dependence of metabolic rate, nutrient supply rate, consumer consumption efficiency and mortality rates all have the potential to affect biomass fluxes between consumers and resources, and in turn, the stability of food webs (Gilbert *et al.* 2014). In plant-herbivore interactions, higher temperature-dependence of heterotroph respiration compared to photosynthesis has been predicted to increase the strength of top down control in aquatic ecosystems (O’Connor *et al.* 2011). However, despite major advances in the ecological theory linking the effects of temperature to metabolism and species interactions (O’Connor *et al.* 2011; Dell *et al.* 2014; Gilbert *et al.* 2014; Amarasekare 2015; Uszko *et al.* 2017), there have been very few empirical tests of this theory, and to our knowledge, no large scale experimental study has confronted recent theoretical developments to test how mismatches in thermal physiology drive the outcome of species interactions.

In aquatic ecosystems, temperature and nutrients are the two main drivers of phytoplankton productivity (Litchman *et al.* 2010). The effects of temperature on phytoplankton growth typically follow a characteristic left-skewed unimodal function, where rates increase exponentially to an optimum followed by a steeper exponential decline (REF). Phytoplankton exhibit substantial variation among species and functional groups in these thermal response curves (Thomas *et al.* 2016) and interspecific variation in thermal tolerance can be an important driver of community dynamics and seasonal succession in phytoplankton communities (Grover & Chrzanowski 2006). Nutrient availability also has a major impact on phytoplankton growth, with rates typically increasing as a saturating, hyperbolic function of increasing nutrients, often characterised by the Monod curve (Monod 1949). Interspecific variation in the functional traits that shape nutrient uptake and growth (e.g., the half saturation constant, which quantifies performance at low nutrient concentrations, and the maximum growth rate in a nutrient saturated medium) are widely recognised to be key drivers of competition (Tilman 1981), community assembly (Bulgakov & Levich 1999) and ultimately the productivity of phytoplankton communities (Behrenfeld *et al.* 2005). The non-linear effects of temperature and nutrients also interact multiplicatively. For example, temperature can influence both the half-saturation constant and the maximum growth rate (Aksnes & Egge 1991; Sterner & Grover 1998; Carter & Lathwell 1967; Mechling & Kilham 1982; Senft *et al.* 1981) and vice-versa, recent work has shown that the optimum temperature for growth increases as a saturating function of nutrient availability (Thomas *et al.* 2017). Thus changes in environmental conditions can potentially amplify mismatches between competitors’ functional traits, and this could affect species competition and community assembly (Litchman *et al.* 2010; Kordas *et al.* 2011). Given that both temperature and nutrient balance are predicted to shift with global changes (IPCC 2013; Behrenfeld *et al.* 2006; Ye *et al.* 2011), understanding the potential for such climate-driven mismatches is made all the more urgent.

Here we investigate how mismatches between species’ traits defining thermal physiology affect competition in phytoplankton. We do so by modelling species growth rate as a temperature dependent Monod model (Monod 1949). Species’ physiology is defined by the two parameters of the model, the maximum growth rate and the half-saturation constant, which are themselves expected to vary with temperature. We assess whether mismatches in the temperature dependence of these traits can predict competitive outcomes. We then test our model’s predictions against empirical data on six species of freshwater phytoplankton in a large-scale experiment with all pairwise combinations of the six species at two temperature and nutrient levels.

# Theory

In line with the experiments (see Methods below), we model the scenario where two species colonise a novel environment at low densities. In this context, we hypothesise that the differences in exponential growth rate (and therefore differences, or mismatches, between species in the traits and variables that give rise to this growth) are key in shaping the competitive outcome between two species. The simplest model of two species competing for a single limiting non-renewable resource undergoing exponential growth can be described by

where is the phytoplankton cell density (cells·mL-1), is the realised exponential growth rate (d-1) of thespecies (subscripts and ), and is time (days). The growth rate of species can in turn be modelled using the Monod equation:

with being the maximum growth rate in nutrient saturated conditions (d-1), the half-saturation constant (μmol·L-1) which is a measure of performance at low nutrient concentrations, and the nutrient (phosphate) concentration in the medium (μmol·L-1). We here assume that remains relatively constant (or is depleted very slowly) throughout an experiment (see Section SX for the rationale and assumptions made by the model). The parameters of the Monod equation, and , can be considered as ‘functional traits’ that characterise a species’ physiology. These traits have been shown to vary among species and play an important role in shaping competitive dynamics in phytoplankton communities (REF). We expect maximum growth rate to be tightly coupled to metabolism, and consequently the temperature dependence of is expected to follow a left-skewed unimodal function of temperature, where rates increase exponentially to an optimum followed by a steeper exponential decline (REF). Within the ‘operational temperature range’ (OTR), the temperature range most likely to be encountered by the population, could be assumed to increase exponentially with temperature (Martin & Huey 2008; Pawar *et al.* 2016). The effects of temperature on are poorly understood and empirical studies have documented a wide range of temperature dependence functions ( Sterner & Grover 1998; Carter & Lathwell 1967; Mechling & Kilham 1982; Senft *et al.* 1981), although a simple theoretical model suggests is expected to increase with temperature (Aksnes & Egge 1991), and another study approximated the temperature dependence of as an exponential function (Reuman *et al.* 2014). Let the temperature dependence within the OTR of both and follow the Boltzmann-Arrhenius equation:

where and are the values of and at a reference temperature (in K), and are the activation energies (eV) that set the relative rate of increase in and with temperature, is the Boltzmann constant (eV·K-1), and is the temperature (K).

Assuming the starting densities are equivalent (), we can define the fitness coefficient of species relative to species by taking the ln ratio of their abundances at time :

(see Section SX). Note that comes about via the differences, or mismatches, in the competing species’ physiology, that is to say, via mismatches in the respective parameters that define the temperature dependence of and (, , , and ). When there are no mismatches (the equivalent parameters are the same in both species), and both species are expected to be equally abundant at any time point . When there are mismatches, , and the sign of indicates which species has a fitness advantage: for , species is expected to outnumber species at time , while the opposite is true for . While we cannot be sure that at time we observe the final competitive outcome between the two species, for simplicity we henceforth refer to the state of a competition (the fitness coefficient) at time as the “competitive outcome”, and refer to the species with greater population size as the “winner” of the competition.

We can assess how important nutrient limitation () is for predicting the competitive outcome by comparing above to a simplified version that assumes nutrient saturation:

In this case, species will grow faster than species *b* if , and therefore if

The trade-off between normalisation constants and activation energies here is explicit: to gain competitive advantage, species can either increase its normalisation constant or activation energy relative to species (see Figure S1 in supplementary information).

The sign of and can change with temperature; this “reversal” in the fitness coefficient indicates that one species could grow faster than another within a specific temperature range (see Section SX and Figure S2).

# Methods

## Study design

We used an experimental approach to test the this simple model’s ability to predict competitive outcomes in a context of climate change, using six species of freshwater phytoplankton. We hypothesise that competition will be driven by mismatches in the temperature dependence of individual species’ physiological traits. We first determined the temperature dependence of and , for each species independently, and then competed the six species in all pairwise combinations at two temperatures and two nutrient levels. The results from the competition experiment were then matched to the predictions made by the theory, using the empirical data on species’ thermal physiology to parametrize the model. Agreement between the model predictions and experimental results would indicate that mismatches between species’ individual traits can largely determine their interaction. Conversely, a low predictive power would suggest that mismatches in nutrient physiology alone are not the sole driver of the competition, and that other factors not included in the model, such as interspecific interference and density dependent growth, could account for the discrepancies (see Section SX).

## Species and culture conditions

The experiment was conducted with six species of freshwater green algae that are known to naturally co-occur, *Ankistrodesmus nannoselene, Chlamydomonas moewusii, Chlorella sorokiniana, Monoraphidium minutum, Scenedesmus obliquus* and *Selenastrum capricornutum* (Fritschie *et al.* 2014). We chose these 6 species because (i) they are similar cell size and (ii) can be cultured on the same media (e.g. standard COMBO culture medium without animal trace elements (Kilham *et al.* 1998)). Strains of each species were ordered in October 2015 from the Culture Collection of Algae and Protozoa ([www.ccap.ac.uk](http://www.ccap.ac.uk), see Supplementary Table 1 for detailed information about the strains). Upon arrival, species were grown on COMBO culture medium, and maintained in semi-continuous culture in an Aralab incubator at 15°C on a 12:12 light-dark cycle with a light intensity of 90 µmol·m-2·s-1.

## Nutrient and temperature dependence of growth rate

We measured growth rates of the 6 species of green algae across gradients in temperature and phosphate concentration. Each of the 6 species was grown in a factorial experiment at 5 temperatures and 13 phosphate concentrations, with 3 replicates per combination, amounting to a total of 1170 cultures. We created 13 solutions of different phosphate concentrations ranging from 0.01 µmol·L-1 of phosphate to 50 µmol·L-1 of phosphate (original phosphate concentration in the COMBO medium) by mixing different amounts of COMBO medium with and without potassium phosphate dibasic (Table S1B). This range was relevant to phosphate concentrations commonly found in lakes (Downing *et al.* 2001). Small tissue culture flasks (Nunclon) filled with 40 mL of each solution were inoculated with each species in monoculture with around 100 cells·mL-1. Samples were diluted or concentrated by filtration to allow for the same inoculation volume, 10 µL (for the very low phosphate concentrations, 0.01, 0.1 and 0.5 µmol·L-1) and 50 µL (for all of the other samples), ensuring that the increase in phosphate concentration due to the inoculum was minimal (respectively 0.01 and 0.06 µmol·L-1). Samples were then grown in Percival incubators at 15, 20, 25, 30, and 35°C on a 12:12 light-dark cycle and with a light intensity of 90 µmol·m-2·s-1 (range: 70-110). Every day, samples were shaken and their position inside of the incubators was randomly changed. Every two days, a 200 µL sample was taken and 10 µL of 1% sorbitol solution was added as a cryoprotectant. After one hour of incubation in the dark, samples were frozen at -80°C until further analysis. Cell density in each sample was determined by flow cytometry (BD Accuri C6). Plates were thawed in a water bath at *ca* 38°C for 10 minutes and then run on the flow cytometer on fast flux settings (66 µL·min-1), counting 10 µL of each sample. Cleaning fluid was run after each species to avoid contamination of measurements between species. The experiment was run for one month. During the experiment, some samples failed to grow properly and were therefore removed from the subsequent analyses.

## Species competition

To investigate the joint effects of temperature and phosphate availability on competitive outcomes among the 6 species of algae, we competed each of the species in all pairwise combinations (15 pairs) at two temperatures (15 and 25°C; low temperature and a temperature close to the optimum for most species) and two phosphate concentrations (saturating [30 µmol·L-1] and limiting [1 µmol·L-1] concentrations, chosen from the Monod curves, see Fig. 1, Fig. S1), each replicated 6 times. We also grew the 6 species in monoculture at the two temperature and nutrient levels. The monoculture trials were divided into two subsets, one training subset, used to train the cell discrimination algorithm, which was replicated 3 times per temperature and nutrient levels and inoculated with 200 cells cells·mL-1, and a testing subset used to test the accuracy of the cell discrimination algorithm, which was replicated 6 times per temperature and phosphate level and inoculated with 100 cells cells·mL-1. In total, the design included 576 samples. The competition experiment was done in twenty-four 24 well plates filled with 2 mL of media, and inoculated with 100 or 200 cells·mL-1 of each species. The position of the species pairs were randomised within the plates, however given the large number of samples and to minimise experimenter error, we separated low-P from high-P plates. Plates were covered with AeraSeal breathable membrane, minimising evaporation and contamination but allowing gas exchange. The competition plates were incubated in the same way as described above for the monoculture growth curves. After 14 days, which was identified from the monoculture experiments as being sufficient time to reach stationary phase, a 200 µL sample was taken and preserved in the same way as described above. Cell density in each sample was determined by flow cytometry (BD Accuri C6) on the slow flux setting (14 µL·min), counting 20 µL of each sample. Cleaning fluid was run after each sample to avoid contamination of measurements between samples.

## Data analyses

All statistical analyses were undertaken using R v3.3.2 (R Core Team 2014).

## *Nutrient and temperature dependence of growth rate*

To characterise the effects of phosphorous availability and temperature on growth we estimated specific growth rate for each of the 1170 combinations of species, phosphate and temperatures from the time-series of cell densities. Population dynamics were fitted to the Buchanan three-phase linear growth model (Buchanan *et al.* 1997) using non-linear least squares regression.

(x)

where is the duration of the lag phase (days), is the time when the maximum population density is reached (days), is the log10 of the initial population density (log10(cells·mL-1)), is the log10 of the maximum population density supported by the environment (log10(cells·mL-1)), and is the specific growth rate (day-1). Fits to the Buchanan model were determined using the ‘nlsLM’ function in the ‘minpack.lm’ package in R (Elzhov *et al.* 2010), which uses the Levenberg-Marquardt optimisation algorithm. Parameter estimation was achieved by running 1000 different random combination of starting parameters picked from uniform distributions and returning the parameter set with the lowest AICc score.

The Monod equation (Eq 1, Monod 1949), was fitted to the estimates of for each species at each temperature and for each of the three replicates using the ‘nlsLM’ function in the ‘minpack.lm’ package. Parameter estimation was achieved by running 1000 different random combination of starting parameters picked from a uniform distribution and returning the parameter set that returned the lowest AICc score.

We used two approaches to describe the thermal variation in and : generalized additive models (GAMs), and Boltzmann-Arrhenius models. For each species, we fitted a GAM model of each parameter with temperature as a smoother term with the number of knots fixed at 3 with the gam function from the mgcv package. We also fitted the Boltzmann-Arrhenius model on linear scales using nonlinear least squares (as above) to the values of and averaged across replicates, for temperatures up to 25°C, and using a reference temperature °C. This analysis produced normalisation constants and activation energies for both and , per species, which we then used to predict the fitness coefficients of equations XX and XX.

***Competition***

FSC files returned by the flow cytometer were read into R using the Bioconductor package ‘FlowCore’, returning side scatter (SSC), forward scatter (FSC), green fluorescence (FL1), orange fluorescence (FL2), red fluorescence (FL3), and blue fluorescence (FL4) values that could be used to define species morphology and thus discriminate between species in pairwise competition samples and determine species identity for each cell. We first filtered the data to remove noise by removing every data point where either ln(FSC.H)<10.3, ln(SSC.H)<3 or ln(FL3.H)<1.5, which are below minimum values observed for life cells of all species. We then separated the data set into 3 data frames, one for the isolates inoculated at 100 cells·mL-1, and one for the isolates inoculated at 200 cells·mL-1, and one for the competing species. The 200 cells·mL-1 isolates dataset measured at day 14 was used to determine pairwise discrimination functions between pair of species. We first removed outliers from this dataset by manually inspecting FSC.H by FL3.H clustering plots and choosing visual thresholds for these two values for each species. We then applied 3 different procedures to discriminate between pairs of species for each temperature and phosphate level: a linear discriminant analysis with the ‘lda’ function from the ‘MASS’ R package, a random forest analysis with the ‘randomForest’ function from the ‘randomForest’ R package and a recursive partitioning and regression tree analysis with the ‘rpart’ function from the ‘rpart’ R package. These analyses were performed using the natural logarithm of the 10 morphological variables returned by the flow cytometer (that is FSC.H, FSC.A, SSC.H, SSC.A, FL1.H, FL1.A, FL2.H, FL2.A, FL3.H, FL3.A, FL4.H and FL4.A, .H standing for height and .A for area), on each of the 15 pairs of species for each combination of temperature and phosphate level. These different discriminant functions were then applied to the 100 cells·mL-1 isolates dataset previously filtered by removing visually determined outliers to test the accuracy of the predictions for the different discriminant methods. We then chose the method that gave the maximum level of accuracy to apply to the competition dataset (Fig. S2A). The best method was the linear discriminant analysis that gave 84 % of accuracy in predicting species identity (Table S2A).

After determining species identity for each sample, we computed cell density and calculated competition coefficients as the proportion of cells from the focal species over the total number of cells. We also computed a binary competition outcome where the competitive dominant was defined as the species that attained more than 50% of the total number of cells.

# Results

## *Nutrient and temperature dependence of growth rate*

The responses of growth rate to the gradients in phosphate concentration were well fit by the Monod equation (Fig. 1a). The half-saturation constant, , and the maximum growth rate, , changed with temperature, and the temperature response of these traits differed between the six species (Table S3A and S3B). Maximum growth rate exhibited a unimodal temperature dependence in *Ankistrodesmus*, *Chlamydomonas*, and *Selenastrum* (Fig 1b, Table S3A). In *Chlorella* and *Monoraphidium*, increased monotonically and did not reach their optima by 35°C, while *Scenedesmus* exhibited negligible temperature dependence (Fig 1b, Table S3A). increased with temperature for *Ankistrodesmus* and *Chlamydomonas*, while *Chlorella* and *Selenastrum* exhibited a unimodal response to temperature and there was no discernible trend for *Monoraphidium* and *Scenedesmus* (Fig. 1c, Table S3B). and were also positively correlated (Pearson *r* = 0.45 [0.27,0.60], t = 4.77, df = 88, p <0.001), highlighting a trade-off between maximum growth rate and performance at low nutrient concentrations.

## *Species competition*

The competitive outcome depended on temperature, nutrient conditions and species pair identity (Fig. 2). For instance, for the pair *Chlorella-Ankistrodesmus*, *Chlorella* dominated the competition at the lower temperature, while *Ankistrodesmus* dominated at the higher temperature under both nutrient conditions. For the pair *Monoraphidium-Chlorella*, *Monoraphidium* won except at high nutrient concentration and high temperature. For the pair *Scenedesmus-Chlamydomonas*, at the lower temperature, there was no clear winner between the two species regardless of nutrient conditions, while at the higher temperature the outcome depended of nutrient conditions: at low nutrient conditions, there was no clear winner while at high nutrient conditions *Chlamydomonas* won (Fig. 2). For some species pairs, the competitive outcome was the same across temperatures and nutrient concentrations. For example, *Chlamydomonas* always won against *Ankistrodesmus*, while *Ankistrodesmus* always beat *Monoraphidium* (while *Monoraphidium* won against *Chlamydomonas* at the higher temperature and lower nutrient concentration).

The fitness coefficient based on individual species’ thermal physiology predicted the correct experimental outcome 68% of times. This result is significant: only 0.9% of random parameter combinations led to a higher predictive power. Results remained largely the same when considering the two nutrient concentrations separately, but competitions at the lower temperature were better predicted by the theory. The predictability of the competitive outcome was also very dependent on the species involved (Table 1). Over 80% of competitive outcomes were correctly predicted for *Ankistrodesmus* and *Chlorella*, while competitions involving *Selenastrum* were considerably more difficult to predict (45%, with almost half the random parameter combinations leading to a greater predictive power). This could, in part, be due to the lesser power of discrimination between cells in pairs involving this species (Table S2A), as well as to the wider confidence intervals around and for this species (Fig 1). Indeed, only removing competitions involving *Selenastrum* increased the overall predictive power to 80%. Assuming nutrient saturated conditions () reduced the overall predictive power to 67% (Table 1); accounting for nutrient uptake barely had an effect on the results. In fact, contrary to expectation, accounting for nutrient uptake did not improve predictions at the lower nutrient concentration. Predictions by subset were very similar to those obtained using ; indeed, for some species, the inclusion of nutrient uptake lowered predictive power (*Ankistrodesmus* and *Chlorella*). Overall, our results clearly show that accounting for nutrient uptake in the fitness coefficient barely yields any improvement in predictive power. The results were robust to the statistical method used to discriminate between species (Supplementary material S7), although for the other methods the theory’s predictive power was broadly marginally lower.

In some cases, the fitness coefficient changed depending on the nutrient concentration and/or temperature. For example, *Chlorella* won against *Chlamydomonas* at 15°C, but the reverse was true at 25°C. These reversals in the fitness coefficient were far more likely to occur between temperatures (in 18 out of 30 competitions; 15 pairs and two nutrient concentrations) than between nutrient concentrations (six out of 30). In the 18 reversals due to the change in temperature, 11 (61%) were correctly predicted by (Tables 1 and 2). Reversals in fitness coefficient were not significantly different between nutrient concentrations or between species. The theory’s ability to predict reversals in the fitness coefficient largely mirrored its ability to predict the fitness coefficients: a greater proportion of reversals were correctly predicted for *Ankistrodesmus* (71%), *Chlorella* (88%), and *Monoraphidium* (75%), but the proportion was low for *Selenastrum* (40%).

# Discussion

Global change is predicted to affect both the temperature of aquatic ecosystems (IPCC 2013) and their nutrient balance (e.g., through an increase in vertical stratifications, reducing nutrient supply (Behrenfeld *et al.* 2006), or through an increase in eutrophication, increasing nutrient supply (Ye *et al.* 2011)). These shifts could lead to mismatches between competing species’ physiological traits, and influence their interactions. Here we showed that phytoplankton species differed in their thermal physiology, and that this variation affected competition between species. Mismatches between species’ temperature dependence of maximum growth rate and half-saturation constant often determined the competitive outcome, and occasionally led to reversals in the fitness coefficient across two different temperatures.

We found that traits governing species’ nutrient physiology varied plastically with temperature.Growth rates of each species depended non-linearly on both temperature and nutrients. Half-saturation constants generally increased with temperature. These results are in agreement with previous results that found a positive relationship between and temperature in plants (Carter & Lathwell 1967) or in algae for nitrogen (Aksnes & Egge 1991; Sterner & Grover 1998) and silicate (Mechling & Kilham 1982), and a hump shaped relationship between and temperature in algae for phosphorus (Senft *et al.* 1981). Studies investigating effects of climate change on algal biomass often consider the half-saturation constant to be independent of temperature (Goldman & Carpenter 1974; Ye *et al.* 2011; Thomas *et al.* 2017); our results highlight the temperature-dependence of nutrient-limited growth. Further, the relationship between temperature and nutrient physiology depended on species identity, with, for instance, distinctly higher half-saturation constants in *Selenastrum* than in *Ankistrodesmus*.

Competitive outcomes between pairs of species varied with temperature and nutrient conditions. These results match previous studies, where temperature has been shown to influence competitive interactions in various groups including phytoplankton, arthropods and vertebrates (see (Dunson & Travis 1991) for a review). Mismatches in the temperature dependence of thermal physiology were a good predictor of competitive outcomes between species. In fact, assuming nutrient saturation () produced almost the same predictive power as incorporating the effects of nutrient limitation (), even at the low nutrient concentration (Table 1), suggesting that it is knowledge of mismatches in the temperature dependence of that is essential in predicting the competitive outcome. This could be due to the relatively large confidence intervals around our estimates of , or to the fact that the lowest concentration of phosphate used in the competition experiment, 1 μmol·L-1, was still relatively high compared to the half-saturating constant of most species. Our results on the predictability of competitive outcomes (Table 1) should also be interpreted in the context that competitions were very variable across replicates (Figure 2), that is to say, both competing species were often observed to win across six replicates. This suggests that there is natural variability in species’ traits, something that is not captured by the model. Despite this variability, competitive outcomes were highly predictable when excluding competitions involving *Selenastrum*, suggesting that the model assumptions are nonetheless appropriate for the other species (and implying that there is little direct interference between them). On the other hand, the predictability of *Selenastrum*’s competitive outcomes was poor. This might have been due to the fact that our discriminating power for this species was low (Table S2Ab), but could also indicate that competitions with this species might have involved some significant form of direct interspecific interference (e.g., through the production of toxins), as also indicated by the fact that competitive interactions involving this species were all strongly negative, leading to a strongly diminished yield of the pair of competitors relative to the same species in monoculture (Loreau & Hector 2001) deviation from expected yield in pairs involving the focal species, mean ± SD, -0.77 ± 0.36 in pairs involving *Selenastrum*, compared to -0.37 ± 0.64, -0.13 ± 0.51, -0.19 ± 0.72 and -0.23 ± 0.53 in pairs involving *Ankistrodesmus*, *Chlamydomonas*, *Chlorella*, *Monoraphidium* and *Scenedesmus* respectively).

We highlighted different competitive interactions where some species dominated at both temperatures and nutrient concentrations, while there were also frequent reversals of competitive outcomes, particularly with temperature and less so with nutrients. Reversals in competitive outcomes were often linked to analogous reversals in the fitness coefficient. Therefore having a better understanding on the thermal-dependency of species’ nutrient physiology is an important step if we are to understand how species competition and community functioning can be affected by climate change (Litchman & Klausmeier 2008; Litchman *et al.* 2010). The results of our study contrast with some earlier studies. For example, (Park 1954) found that higher growth rate of a competitor at higher temperature did not lead to a switch in competitive dominance in *Tribolium* species.

More generally, our findings stress the importance of considering how species traits will plastically change with temperature to better understand biotic interactions in a context of global climate change. Studying consequences of climate change in terms of mismatches between the temperature dependence of physiological traits should be a useful approach in understanding how species interactions will be modified by warming climates (Dell *et al.* 2014). Further, because global changes are unlikely to act only through temperature changes but should involve rapid modifications of both nutrient and thermal conditions (Behrenfeld *et al.* 2006; Ye *et al.* 2011; IPCC 2013), it is crucial to better understand how the combination of multiple stressors should affect species and community responses to global changes. We highlight the interest of considering a spectrum of different ecological contexts to predict successful competitors and invaders, and to pinpoint which mismatch in species traits is more important in which ecological context.

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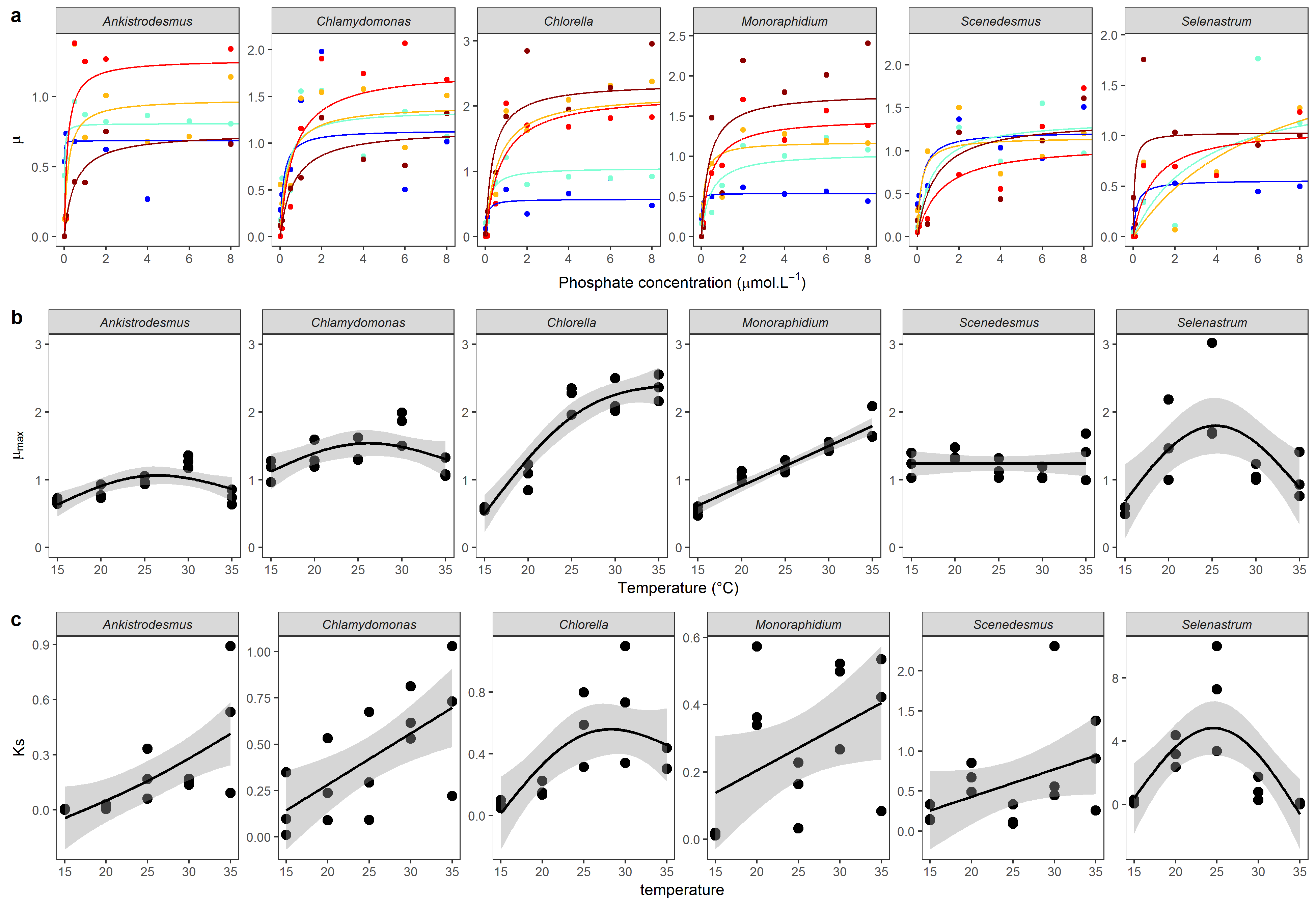
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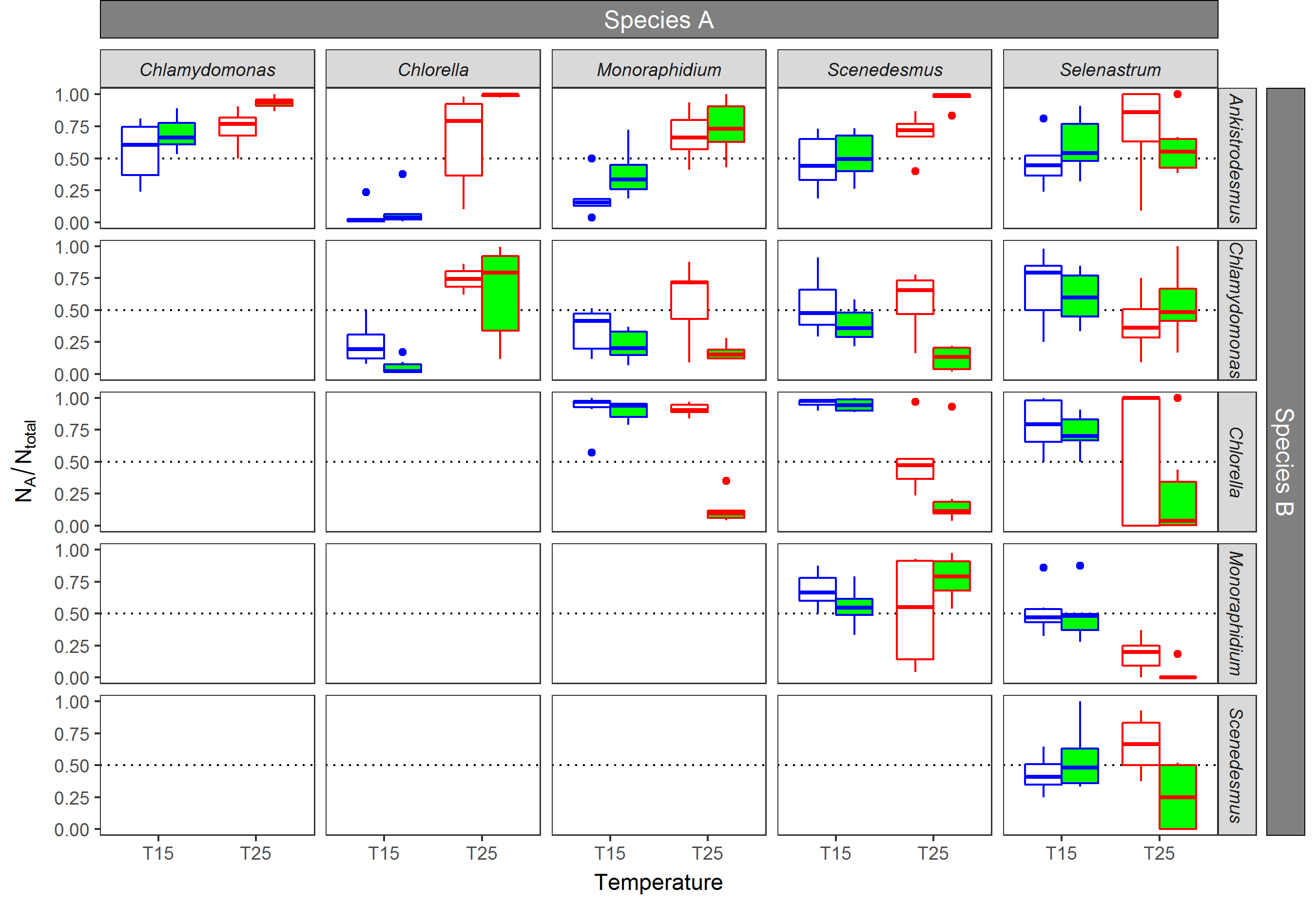
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# Figures:



## **Fig 1**:

**(a)** Mean Monod curves for each species growth rate. Growth rate as a function of phosphate concentration in the medium (μmol·L-1) and temperature (from blue: 15°C to dark red: 35°C). Points represent the mean of the 3 replicates, and the Monod curve is drawn from the mean of the rate and parameters from the 3 replicates. Note that the phosphate concentration levels in the experiment go from 0.01 to 50 μmol·L-1 but the x-axis was cut at 8 μmol·L-1 for clarity. (**b)** Maximum growth rate as a function of temperature. **c**Lines represent the fit of the GAM models investigating the temperature dependence of each parameter. See Tables S3A and S3B for more details about the temperature-dependence of the estimates from the Monod model.



## Fig. 2:

Competition between species. For each pair of species, the proportion of cells from species A over the total number of cells at day 14. Colours represent the temperature of the trial, blue: 15°C, red: 25°C; and fills represent the nutrient conditions of the trial, white: non-saturated nutrient solution (1 μmol·L-1 of phosphate), green, saturated nutrient solution (30 μmol·L-1 of phosphate). Boxplots represent the values of the 6 replicates per condition. The dotted line represents the situation where 50% of the total number of cells pertain to the species A.

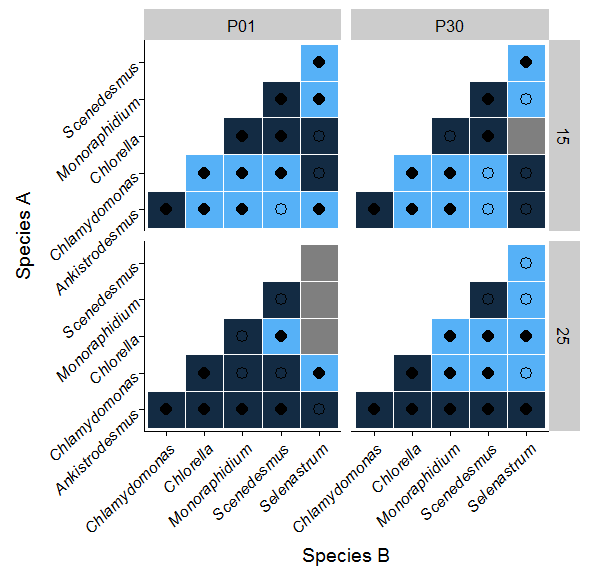


Figure 4: Outcome of the competition and validity of the model predictions for each pair of species depending on the temperature and phosphate level. The colour indicates the identity of the winning species (light blue species A wins, dark blue species B wins), and the shape inside shows whether the model prediction is correct (filled, the model is correct, empty, the model is incorrect).

# Tables

**Table 1**: Proportion of competition outcomes correctly predicted by theory. Results are shown for the full dataset (including competitions at both temperatures and nutrient concentrations), by temperature, by nutrient concentration, and by species (where only competitions involving each individual species is considered in turn). The column “” uses eqn. X in the main text, and assumes nutrient saturated conditions, while “” corresponds to eqn. X in the main text, and explicitly captures nutrient limitation. “N” indicates the number of competitions in each subset. Column “ reversals” shows the proportion of competitions for which a reversal in the competitive outcome was observed between temperatures that was correctly predicted by the theory. For example, a value of 0.5 here means that the theory correctly predicted 50% of the observed reversals in the competitive outcome (given by column “N revs.”). P values indicated in parentheses were obtained by bootstrapping (see Section SX in supplementary information). The experimental competition data uses the LDA discrimination method. Analogous results for the random forest and rpart discrimination methods are shown in Tables XX and XX.

|  |  | |  | | **N** | **reversals** | | **N revs.** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *Full dataset* | | | | | | | | |
|  | 0.67 | (0.013) | 0.68 | (0.009) | 60 | 0.61 | (0.001) | 18 |
| *By temperature* | | | | | | | | |
| °C | 0.70 | (0.045) | 0.73 | (0.032) | 30 |  |  |  |
| °C | 0.63 | (0.122) | 0.63 | (0.125) | 30 |  |  |  |
| *By nutrient* | | | | | | | | |
| [P] = 1 µmol·L-1 | 0.67 | (0.013) | 0.67 | (0.018) | 30 | 0.60 | (0.001) | 10 |
| [P] = 30 µmol·L-1 | 0.67 | (0.035) | 0.70 | (0.018) | 30 | 0.62 | (0.001) | 8 |
| *By species* | | | | | | | | |
| *Ankistrodesmus* | 0.85 | (0.004) | 0.80 | (0.011) | 20 | 0.71 | (0.011) | 7 |
| *Chlamydomonas* | 0.60 | (0.177) | 0.70 | (0.058) | 20 | 0.33 | (0.061) | 6 |
| *Chlorella* | 0.90 | (0.000) | 0.85 | (0.007) | 20 | 0.88 | (0.002) | 8 |
| *Monoraphidium* | 0.60 | (0.092) | 0.70 | (0.031) | 20 | 0.75 | (0.000) | 4 |
| *Scenedesmus* | 0.60 | (0.074) | 0.60 | (0.126) | 20 | 0.50 | (0.003) | 6 |
| *Selenastrum* | 0.45 | (0.469) | 0.45 | (0.565) | 20 | 0.40 | (0.034) | 5 |



### **Table2:**

Predictability of reversals in competitive outcomes across the two experimental temperatures. . N values indicate total numbers in each category, while the 2x2 matrix shows in how many of those cases experiments and theory (assuming nutrient saturation) coincide. For example, the number 11 indicates that 11 out of the 18 reversals observed in experiments were correctly predicted by theory (while a total of 20 reversals were predicted by the theory).

|  |  |  |
| --- | --- | --- |
|  | **Reversal in** | |
| **Reversal in competition** | Yes (N = 20) | No (N = 10) |
| Yes (N = 18) | 11 | 6 |
| No (N = 12) | 4 | 4 |

# Supplementary Information

## S1: Theory

We aim to quantify competitive advantage or relative fitness of the competing phytoplankton species in a potentially nutrient limited environment, and how physiological mismatches affect this competitive advantage. Our starting point follows Tilman’s (1977, 1981) model of two phytoplankton populations competing for a single limiting nutrient () in a chemostat-type environment:

Here, is the -th species density (cells·mL−1), is its realised growth rate (d−1), is its maximum growth rate in nutrient saturated conditions (d−1), is the half-saturation constant (μmol·L−1) (the nutrient concentration at which realised growth is ; a measure of performance at low nutrient concentrations), is the nutrient concentration (μmol·L−1), is dilution rate, and is the inflow concentration of nutrients. The constant converts units of nutrient to phytoplankton cell units (1000·μmol·cell−1); that is, it is the yield of species with respect to the limiting resource (inverse of number of phytoplankton cells produced per unit of resource).

Because the colonisation experiments do not replenish nutrients, , leaving

If we were to assume that nutrients do not deplete significantly over the course of a (14 day) experiment (a reasonable simplifying assumption; see Section SX), the model reduces to

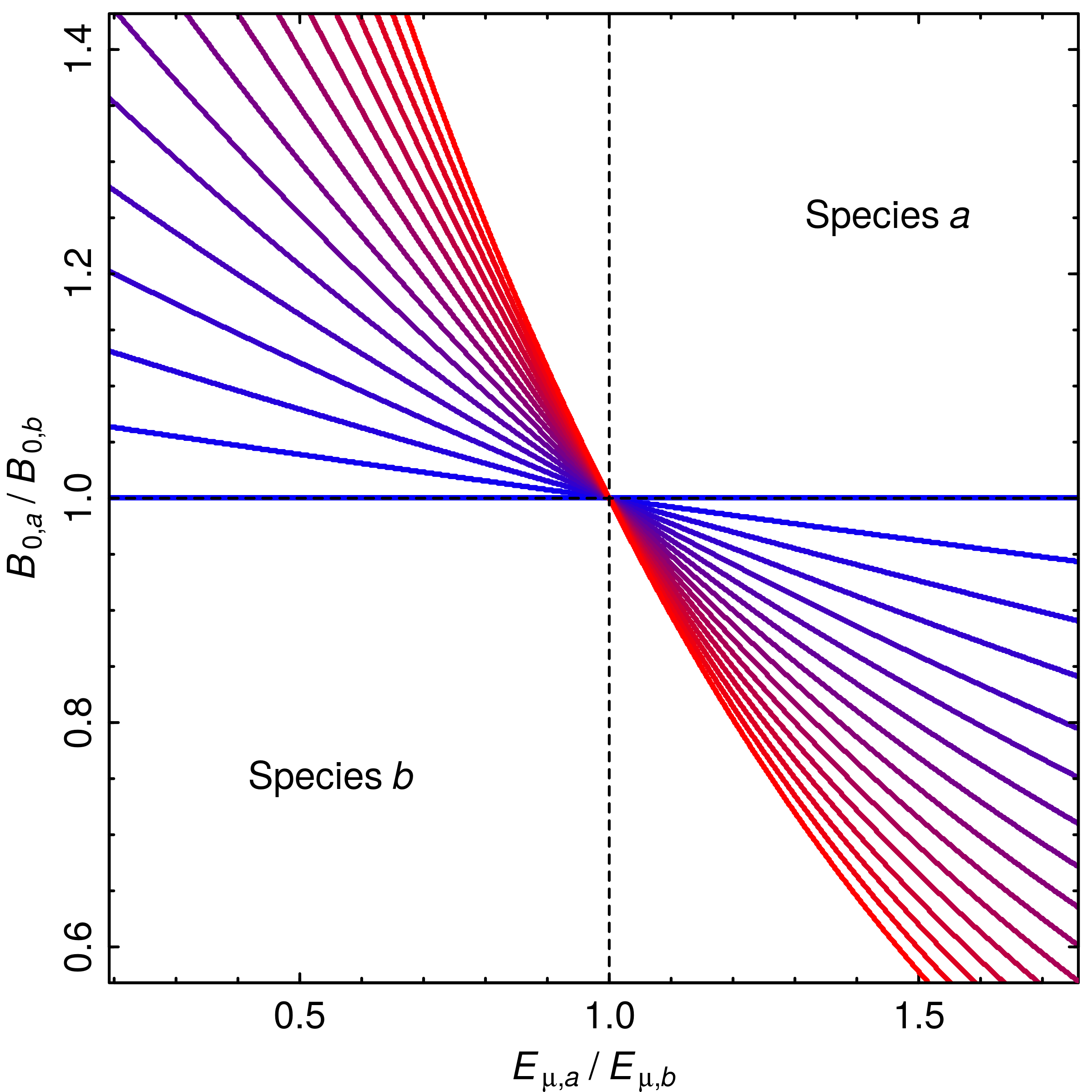
Experiments stipulate the same (small) number of cells at the start of a colonisation experiment, and then estimate the number of cells of each species present after a period of 14 days. The model above can be solved to provide the same information:

where is time (in days) and where the growth rate is modelled using the Monod equation, as above:

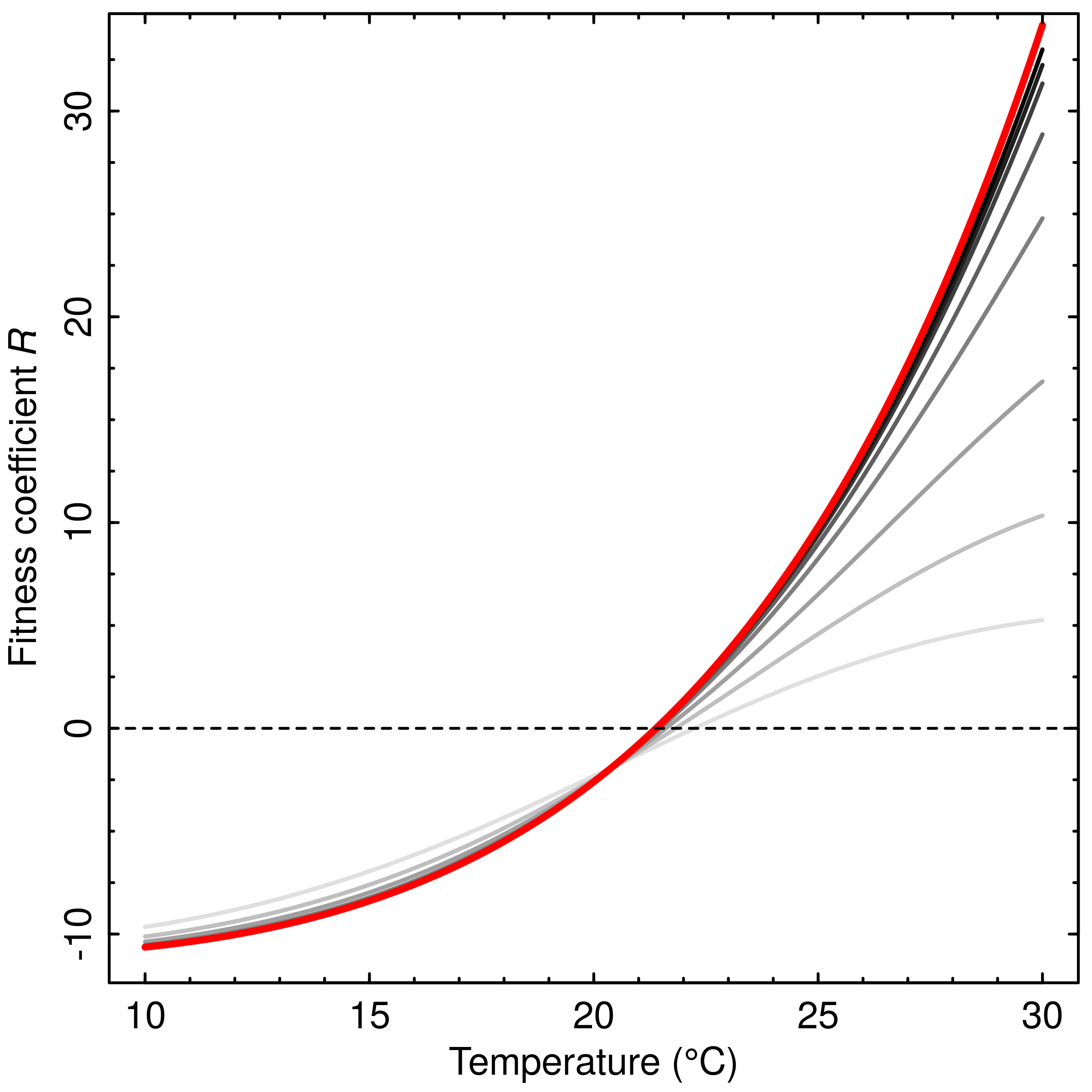
This model is the simplest we can use to model the temperature and nutrient dependent competition of two phytoplankton species, and importantly only incorporates variables measured in experiments. As a result, the model makes a number of simplifying assumptions. We assume that the exponential growth phase in a colonisation is key in determining competitive outcomes, and therefore, comparing exponential growth rates between species competing for a single limiting resource is important for predicting which species might be expected to dominate. However, in the long run, and once populations reach high enough population densities, density dependence and intraspecific competition might be expected to play a role of increasing importance. At higher population densities, competing populations might also be more likely to directly interfere with each other through, for example, through the production of toxins or competition for light (inter-specific competition). Populations are also expected to experience a mortality rate, not captured in the model above. Our focus is on the initial phase of colonisation, so assume most of these factors to be of limited importance in this context. Furthermore, many of these factors are not measured in experiments, and some mechanisms (such as density dependence or intraspecific competition) can be approached a number of different ways; including these would substantially expand the study beyond its scope.

Once and are expressed in terms of temperature (eqns. XX and XX in the main text), a reversal in the fitness coefficient (a change in its sign) can be found numerically. For the nutrient saturated case, the temperature at which is given by

and which species grows faster is determined by the difference in activation energies alone: if , species is expected to outcompete for .



**Figure S1:** Contour lines illustrating the competitive outcome for a range of parameter combinations, assuming nutrient saturation (). The colour of the lines correspond to different temperatures, ranging from 15°C for the blue line, to 30°C for the red line. For example, for = 1 and = 0.8, species *b* grows faster than species *a*, but for = 0.5 and = 1.2, which species grows faster depends on the temperature. Here, , , and °C, meaning that at °C, which species wins is determined by .



**Figure S2:** Example of a reversal in the fitness coefficient across a temperature range. The red line is for nutrient saturated conditions (), and the grayscale lines are for different nutrient concentrations, ranging from µmol·L-1 for the light gray line, to 50 µmol·L-1 for the black line. The example uses parameters for *Chlorella* and *Chlamydomonas*, where means *Chlorella* outcompetes *Chlamydomonas*.

## S1: Experimental design

### Figure S1A: Flow chart of the experimental design

6 phytoplankton species

25°C

15°C

*Ks1*

*μmax*

*μ*

*Ks1*

[P]*1*

35°C

30°C

25°C

20°C

15°C

Growth rate

Nutrient physiology traits

Isolates 🡪Species discrimination function 🡪Applied to pairs of species 🡪Species identity

**Competition coefficients**

**Model**

Input:

Mismatches between traits

Output:

**Competition coefficients**

**Acclimated physiology**

For each species

25°C

Pair of species

Isolate

**Competition experiment**

For each pair of species

****

15°C

Acclimated photosynthesis

*μmax*

**Temperature-and-nutrient-dependent growth rate**

For each species

### Table S1A:

Detailed information about the six species

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Species name** | **Class** | **Order** | **Strain** | **Origin** |
| *Ankistrodesmus nannoselene*  Skuja (1948) | Chlorophyceae | Sphaeropleales | CCAP 202/6A | Siggeforsajon, Sweden |
| *Chlamydomonas moewusii*  Gerlof (1940) | Chlorophyceae | Chlamydomonadales | CCAP 11/5A | Freshwater |
| *Chlorella sorokiniana*  Shihira & Krauss (1965) | Trebouxiophyceae | Chlorellales | CCAP 211/8K | Austin, Texas, USA |
| *Monoraphidium minutum* (Nägeli)  Komarkova-Legnerova (1969) | Chlorophyceae | Sphaeropleales | CCAP 278/3 | Texas, USA |
| *Scenedesmus obliquus* (Turpin) Kützing (1833) | Chlorophyceae | Sphaeropleales | CCAP 276/3B | Lund, Sweden |
| *Selenastrum capricornutum*  Printz (1913) | Chlorophyceae | Sphaeropleales | CCAP 278/4 | Akershus, Norway |

### Table S2A:

Phosphate concentration levels for each solution in µmol·L-1 and µg·L-1. We created 13 solutions of different phosphate concentrations ranging from 0.01 µmol·L-1 of phosphate to 50 µmol·L-1 of phosphate by mixing different amounts of COMBO medium without potassium phosphate dibasic (P- COMBO) and normal COMBO medium (P+ COMBO) in 40 mL nunclons. We used a modified version of the standard COMBO medium without animal trace solution in which we increased the fraction of carbonate by adding 10 mL of a stock solution of 55.8 g·L-1 of sodium bicarbonate to maintain a DIC of more than 6.6 mmol·L-1 in order to prevent carbon limitation, which allowed a C:N:P ratio of 132:20:1 in the P+ COMBO solution, above the Redfield ratio of 106:16:1.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Phosphate concentration (µmol·L-1 ) | 50 | 40 | 30 | 20 | 10 | 8 | 6 | 4 | 2 | 1 | 0.5 | 0.1 | 0.01 |
| Phosphate concentration (µg·L-1 ) | 4750 | 3800 | 2850 | 1900 | 950 | 760 | 570 | 380 | 190 | 95 | 47.5 | 9.5 | 0.95 |
| Amount of P+ COMBO (mL) | 40 | 32 | 24 | 16 | 8 | 6.4 | 4.8 | 3.2 | 1.6 | 0.8 | 0.4 | 0.08 | 0.008 |
| Amount of P- COMBO (mL) | 0 | 8 | 16 | 24 | 32 | 33.6 | 35.2 | 36.8 | 38.4 | 39.2 | 39.6 | 40 | 40 |

## S2: Discrimination between species in the competition experiment

### Table S2A:

A: Proportion of correct assignations for each discrimination method (LDA: linear discriminant analysis, Random Forest analysis, RPART: recursive partitioning and regression tree) summarised by phosphate and nutrient conditions for all pair of species. B: Proportion of correct assignations for each discrimination method summarised by pair of species for all nutrient and thermal conditions. C: Proportion of correct assignations for each discrimination method summarised by species for all nutrient and thermal conditions

A

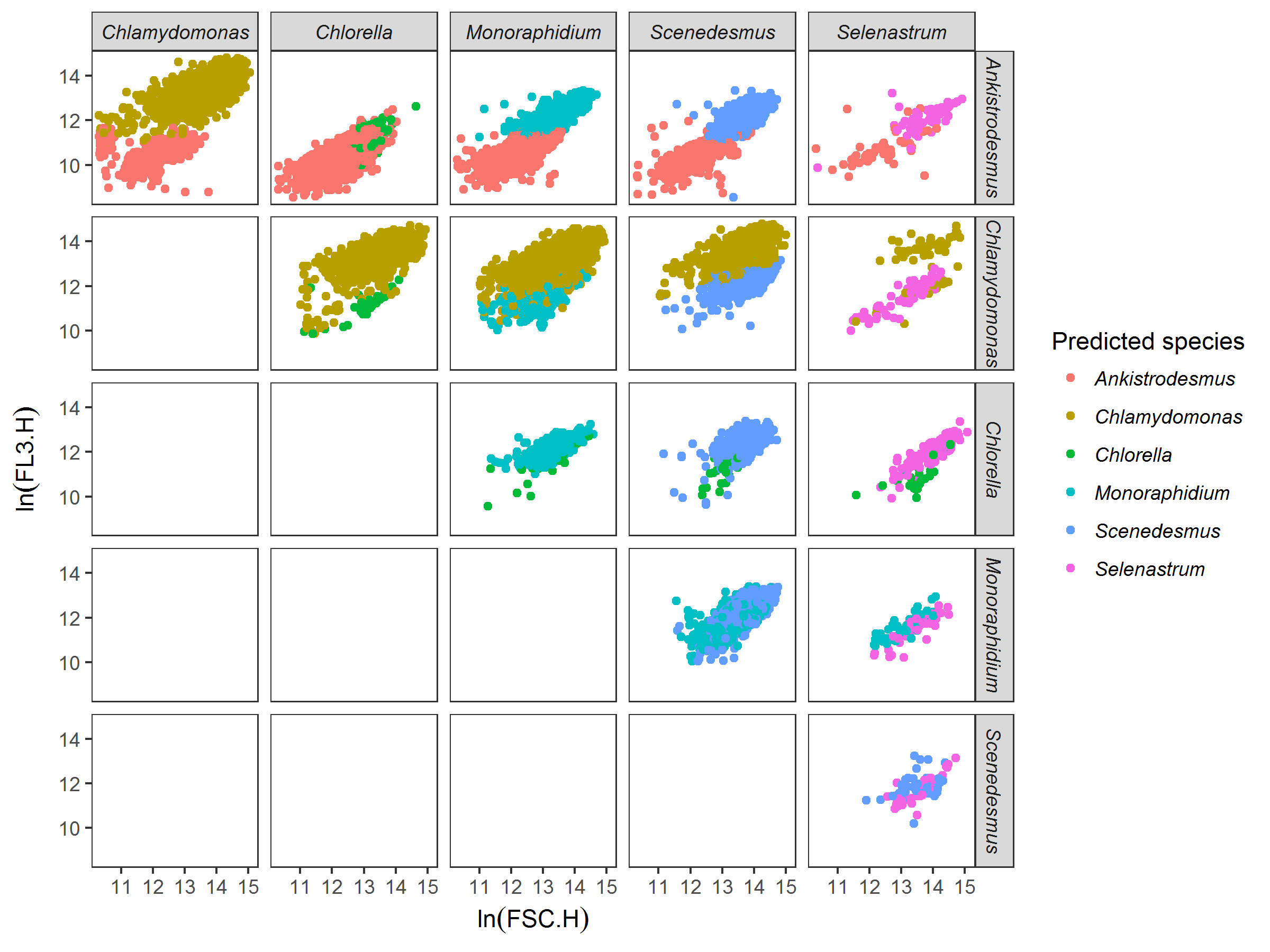
|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Temperature** | **Nutrient** | **LDA** | **Random forest** | **RPART** |
| 15 | 1 | 0.79 | 0.68 | 0.64 |
| 15 | 30 | 0.85 | 0.8 | 0.76 |
| 25 | 1 | 0.7 | 0.69 | 0.68 |
| 25 | 30 | 0.64 | 0.66 | 0.62 |
| **Mean** | | **0.75** | **0.71** | **0.68** |

B

|  |  |  |  |
| --- | --- | --- | --- |
| **Species** | **LDA** | **Random forest** | **RPART** |
| *Ankistrodesmus* | 0.91 | 0.86 | 0.72 |
| *Chlamydomonas* | 0.93 | 0.93 | 0.81 |
| *Chlorella* | 0.85 | 0.86 | 0.67 |
| *Monoraphidium* | 0.84 | 0.78 | 0.65 |
| *Scenedesmus* | 0.83 | 0.77 | 0.61 |
| *Selenastrum* | 0.70 | 0.68 | 0.48 |
| **Mean** | **0.84** | **0.81** | **0.66** |

C

|  |  |  |  |
| --- | --- | --- | --- |
| **Pair of species** | **LDA** | **Random forest** | **RPART** |
| *Ankistrodesmus-Chlamydomonas* | 1 | 1 | 0.94 |
| *Ankistrodesmus-Chlorella* | 0.91 | 0.88 | 0.73 |
| *Ankistrodesmus-Monoraphidium* | 0.87 | 0.74 | 0.71 |
| *Ankistrodesmus-Scenedesmus* | 0.95 | 0.93 | 0.71 |
| *Ankistrodesmus-Selenastrum* | 0.82 | 0.73 | 0.52 |
| *Chlamydomonas-Chlorella* | 0.96 | 0.96 | 0.79 |
| *Chlamydomonas-Monoraphidium* | 0.96 | 0.97 | 0.86 |
| *Chlamydomonas-Scenedesmus* | 0.94 | 0.92 | 0.74 |
| *Chlamydomonas-Selenastrum* | 0.78 | 0.8 | 0.74 |
| *Chlorella-Monoraphidium* | 0.83 | 0.85 | 0.7 |
| *Chlorella-Scenedesmus* | 0.86 | 0.84 | 0.65 |
| *Chlorella-Selenastrum* | 0.67 | 0.76 | 0.48 |
| *Monoraphidium-Scenedesmus* | 0.88 | 0.69 | 0.63 |
| *Monoraphidium-Selenastrum* | 0.68 | 0.67 | 0.33 |
| *Scenedesmus-Selenastrum* | 0.53 | 0.46 | 0.34 |
| **Mean** | **0.84** | **0.81** | **0.66** |



### Figure S2A:

Example of discrimination between species among pairs of species, here for species grown at 15°C in saturating nutrient conditions after 14 days of experiment. Each dot represents a cell, here mapped on FSC.H (size proxy) and FL3.H (chlorophyll a proxy) characteristics from the flow cytometer. Colours represent the species predicted by the discrimination algorithm. The discrimination algorithm is a linear discriminant analysis trained with flow cytometer data (FSC.H, FSC.A, SSC.H, SSC.A, FL1.H, FL1.A, FL2.H, FL2.A, FL3.H, FL3.A, FL4.H, and FL4.A) from the species grown in isolates at the same temperature and nutrient conditions. For example, *Chlamydomonas* outcompetes *Chlorella* in these nutrient and temperature conditions.

## S3: Temperature dependence of the Monod model parameters

### Table S3A:

Results from the GAMs where is a function of temperature, for each species. See Fig 1 for the representation of the GAMs.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Species** | **edf** | **F** | **p-value** | **R2** |
| *Ankistrodesmus* | 2 | 6.74 | 0.011\* | 0.45 |
| *Chlamydomonas* | 2 | 3.42 | 0.066. | 0.26 |
| *Chlorella* | 2 | 54.02 | >0.001\*\*\* | 0.88 |
| *Monoraphidium* | 2 | 63.67 | >0.001\*\*\* | 0.90 |
| *Scenedesmus* | 2 | 0.41 | 0.674 | -0.09 |
| *Selenastrum* | 2 | 5.82 | 0.017\* | 0.41 |

### Table S3B:

Results from the GAMs where is a function of temperature, for each species. See Fig 1 for the representation of the GAMs.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Species** | **edf** | **F** | **p-value** | **R2** |
| *Ankistrodesmus* | 2 | 6.29 | 0.013\* | 0.43 |
| *Chlamydomonas* | 2 | 4.53 | 0.034\* | 0.34 |
| *Chlorella* | 2 | 6.37 | 0.013\* | 0.43 |
| *Monoraphidium* | 2 | 2.17 | 0.157 | 0.14 |
| *Scenedesmus* | 2 | 1.32 | 0.302 | 0.04 |
| *Selenastrum* | 2 | 7.92 | 0.006\*\* | 0.50 |

## S5: Significance of competitive outcomes predicted by the model

To quantify the significance of the theory’s ability to predict competitive outcomes, we ran the analysis 10,000 times, sampling the values of , , , and independently, with replacement, from the pool of available values. When assessing the theory’s performance for a particular subset, for example, for competitions at °C, parameters were sampled independently from all values estimated at °C only. The analysis produced 10,000 proportions of competitive outcomes correctly predicted. The proportion of runs that correctly predicted a greater number of competitive outcomes than the real parameter values are then given as the P values in Table 1. Therefore, P=0.05 means that 500 out of 10,000 random parameter combinations correctly predicted a greater proportion of competitive outcomes. Similarly, the analysis produced 10,000 predictions of reversals in fitness coefficients. The proportions of runs that correctly predicted a greater number of fitness coefficient reversals than the real parameter values are given as the P values in Table 1.

## S6: Robustness of the results to different statistical methods

Estimates for and for the Monod model were obtained from a non-linear mixed model approach with the ‘nlme’ function in R, and were then used in models investigating competitive outcome throughout the manuscript. To test the robustness of the model to the method of determination of and , we also fitted the Monod model to each species and temperature level using the ‘nlsLM’ function in the ‘minpack.lm’ package. Parameter estimation was achieved by running 1000 different random combination of starting parameters picked from a uniform distribution and returning the parameter set that returned the lowest AIC score. The two modelling approaches gave concordant results (Fig S7A). For this reason, we used fits from this latter approach to feed into the theory (Table S7A). The results were extremely similar, with a slightly higher variance on the effect of , which did not affect the predictive power of the model overall.

A second source of uncertainty was due to the method of discrimination between species. We used three different methods of discrimination, a linear discriminant analysis, a random forest analysis and a recursive partitioning and regression tree (rpart, see Supplementary Material S2). Because the linear discriminant analysis was found to have the best predictive power overall (Table S2A), we used this method throughout the manuscript. However, we tested whether our results were robust to the method of species discrimination by comparing results from the competition model to predictions using the random forest analysis and the rpart discrimination method, first with the mixed effect parameters from the Monod model (Table S7B and S7D), and second with the Monod parameters estimated using nonlinear least squares (Table S7C and S7E). The results were similar, with a lower predictive power of each variable and of the model due to the lower discrimination power of the two methods, but no significant discrepancies between species and temperature and nutrient conditions.

### Table S6A:

Same as Table 1 in the main text, using the random forest discrimination method for the competition data.

















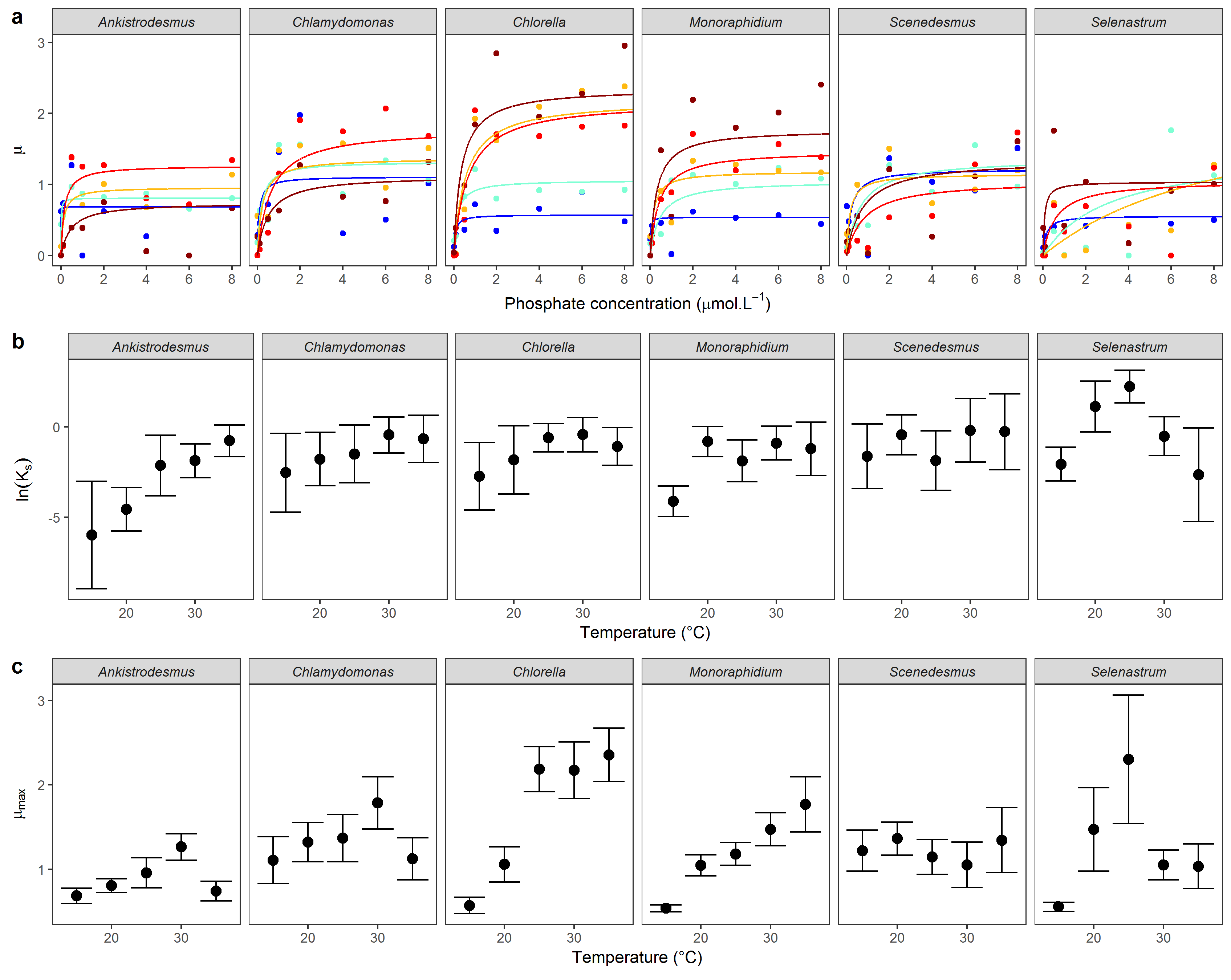


|  |  | |  | | **N** | **reversals** | | **N revs.** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *Full dataset* | | | | | | | | |
|  | 0.63 | (0.043) | 0.72 | (0.004) | 60 | 0.56 | (0.006) | 16 |
| *By temperature* | | | | | | | | |
| °C | 0.70 | (0.041) | 0.73 | (0.030) | 30 |  |  |  |
| °C | 0.57 | (0.276) | 0.70 | (0.062) | 30 |  |  |  |
| *By nutrient* | | | | | | | | |
| [P] = 1 µmol·L-1 | 0.57 | (0.212) | 0.70 | (0.024) | 30 | 0.44 | (0.014) | 9 |
| [P] = 30 µmol·L-1 | 0.70 | (0.008) | 0.73 | (0.004) | 30 | 0.71 | (0.001) | 7 |
| *By species* | | | | | | | | |
| *Ankistrodesmus* | 0.85 | (0.003) | 0.80 | (0.012) | 20 | 0.71 | (0.012) | 7 |
| *Chlamydomonas* | 0.65 | (0.060) | 0.75 | (0.017) | 20 | 0.40 | (0.061) | 5 |
| *Chlorella* | 0.80 | (0.018) | 0.85 | (0.007) | 20 | 0.75 | (0.010) | 8 |
| *Monoraphidium* | 0.60 | (0.098) | 0.70 | (0.030) | 20 | 0.75 | (0.000) | 4 |
| *Scenedesmus* | 0.50 | (0.382) | 0.60 | (0.229) | 20 | 0.25 | (0.115) | 4 |
| *Selenastrum* | 0.40 | (0.604) | 0.60 | (0.160) | 20 | 0.25 | (0.115) | 4 |

### Table S6B:

Same as Table 1 in the main text, using the rpart discrimination method for the competition data.

|  |  | |  | | **N** | **reversals** | | **N revs.** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *Full dataset* | | | | | | | | |
|  | 0.60 | (0.066) | 0.65 | (0.022) | 60 | 0.50 | (0.004) | 18 |
| *By temperature* | | | | | | | | |
| °C | 0.67 | (0.058) | 0.70 | (0.047) | 30 |  |  |  |
| °C | 0.53 | (0.321) | 0.60 | (0.187) | 30 |  |  |  |
| *By nutrient* | | | | | | | | |
| [P] = 1 µmol·L-1 | 0.57 | (0.199) | 0.63 | (0.075) | 30 | 0.44 | (0.010) | 9 |
| [P] = 30 µmol·L-1 | 0.63 | (0.062) | 0.67 | (0.036) | 30 | 0.56 | (0.004) | 9 |
| *By species* | | | | | | | | |
| *Ankistrodesmus* | 0.80 | (0.003) | 0.75 | (0.016) | 20 | 0.62 | (0.010) | 8 |
| *Chlamydomonas* | 0.55 | (0.230) | 0.65 | (0.090) | 20 | 0.29 | (0.079) | 7 |
| *Chlorella* | 0.85 | (0.000) | 0.80 | (0.014) | 20 | 0.86 | (0.002) | 7 |
| *Monoraphidium* | 0.55 | (0.132) | 0.65 | (0.043) | 20 | 0.60 | (0.000) | 5 |
| *Scenedesmus* | 0.45 | (0.465) | 0.55 | (0.293) | 20 | 0.20 | (0.146) | 5 |
| *Selenastrum* | 0.40 | (0.599) | 0.50 | (0.421) | 20 | 0.25 | (0.120) | 4 |

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### **Fig S6A**:

**(a)** Mean Monod curves for each species growth rate estimated using nonlinear least squares. Growth rate as a function of phosphate concentration in the medium (μmol·L-1) and temperature (from blue: 15°C to dark red: 35°C). Points represent the mean of the 3 replicates. Note that the phosphate concentration levels in the experiment go from 0.01 to 50 μmol·L-1 but the x-axis was cut at 8 μmol·L-1 for clarity. **(b)** Half-saturation coefficient (mean ± 95%CI) **(c)** Maximum growth rate (mean ± 95%CI).