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- of diatom cell walls with implications for taxonomy. In Wetherbee, R., Andersen, R. A. & Pickett-Heaps, J. D. [Eds.] *The Protistan Cell Surface*. Springer-Verlag, New York, pp. 43–60.
- Stevenson, R. J. & Peterson, C. G. 1989. Variation in benthic diatom (Bacillariophyceae) immigration with habitat characteristics and cell morphology. *J. Phycol.* 25:120–9.
- Stevenson, R. J., Peterson, C. G., Kirschtel, D. B., King, C. C. & Tuchman, N. C. 1991. Density-dependent growth, ecological strategies, and effects of nutrients and shading on benthic diatom succession in streams. *J. Phycol.* 27:59–69.
- Tanaka, N. 1986. Adhesive strength of epiphytic diatoms on various seaweeds. *Bull. Jap. Soc. Sci. Fish.* 52:817–21.
- Webster, D. R., Cooksey, K. E. & Rubin, R. W. 1985. An investigation of the involvement of cytoskeletal structures and secretion in gliding motility of the marine diatom, *Amphora coffaeiformis*. *Cell Motil.* 5:103–22.
- Werner, D., Ed. 1977. *The Biology of Diatoms*. University of California Press, Berkeley, 498 pp.

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COMPETITION FOR PHOSPHORUS AMONG PLANKTONIC DESMID SPECIES IN CONTINUOUS-FLOW CULTURE¹

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ABSTRACT

When grown under stringent P limitation, affinity for P uptake and growth in *Staurastrum pingue* Teil. and *Staurastrum chaetoceras* (Schr.) G. M. Smith (both originating from eutrophic lakes) were of the same magnitude, whereas these parameters for *Cosmarium abbreviatum* Rac. var. *planctonicum* W. & G. S. West (isolated from a meso-oligotrophic lake) were significantly higher in value. On the other hand, at all growth rates tested, maximum P uptake rates were lower in *C. abbreviatum* than in the two *Staurastrum* species. The outcome of competition between either *Staurastrum* species and *C. abbreviatum* in mixed chemostats under different levels of continuous P limitation was in agreement with what could be predicted from the species-specific affinity parameters: *Staurastrum* was outcompeted at dilution rates lower than 0.012 h^{-1} , calculated to correspond with external inorganic P concentrations lower than $0.02\text{ }\mu\text{M P}$, but won out at higher dilution rates. When P was added in two pulses of $2.5\text{ }\mu\text{mol}\cdot\text{L}^{-1}$ a week instead of continuously, *S. chaetoceras* outcompeted *C. abbreviatum* at a slow rate. When P was supplied as a daily pulse of $0.7\text{ }\mu\text{mol}\cdot\text{L}^{-1}$, a stable co-existence of *S. chaetoceras* and *C. abbreviatum* was established, *Staurastrum* predominating the culture in cell numbers at ca. 85%. The results show that P uptake and growth characteristics of the three species can predict the outcome of competition under various P-limited conditions. Specific growth kinetic parameters as found in this study may also explain distribution patterns of the species observed in the field.

Key index words: Chlorophyta; competition; continuous culture; *Cosmarium abbreviatum*; desmids; ecophysiology; phosphorus limitation; phytoplankton; pulsed culture; *Staurastrum chaetoceras*; *Staurastrum pingue*

Natural selection is thought to favor those algae whose nutrient uptake and growth characteristics are best suited to environmental conditions, so that species originating from habitats differing in trophic state will exhibit different affinities for growth and nutrient uptake kinetics (Hecky and Kilham 1974, Tilman 1977, P. Kilham and Tilman 1979). Consequently, nutrient uptake and growth kinetics may help to explain the distribution of algal species in the field.

Since 1980s a lot of experimental documentation has been presented concerning the importance of nutrient uptake and growth kinetic parameters in determining the outcome of phytoplankton competition (e.g. Holm and Armstrong 1981, Tilman 1981, S. S. Kilham 1984, Healey and Hendzel 1988, Grover 1989a, b, 1991, Olsen et al. 1989, Nicklisch et al. 1991). Yet, what was stressed by Tilman et al. (1982), that is, the need for laboratory studies designed specifically to test the validity of conclusions derived from correlation analyses in the field, still holds. Remarkably enough, none of the experimental studies focused on ecophysiological differences among species whose distribution in the field may be linked to the trophic state of the habitat. Most of the preceding experimental studies include species isolated from the same lake. In addition, experiments were often carried out with species belonging to different algal groups (e.g. cyanobacteria, diatoms, green algae) so that physiological differences observed may be inherent to the groups in question rather than specific to the species level.

Coesel and Wardenaar (1990) determined growth rates of 12 planktonic desmid species in relation to temperature and irradiance. They found significantly lower maximum growth rates in species characteristic of oligotrophic lakes as compared to those that peak in eutrophic ones. This potential disad-

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TABLE 1. Mean cellular dry weight, protein content, and protein: dry weight ratio of *C. abbreviatum*, *S. pingue*, and *S. chaetoceras* at different steady-state growth rates. Mean values $\pm$ SD.

μ (h ⁻¹)	<i>C. abbreviatum</i>			<i>S. pingue</i>			<i>S. chaetoceras</i>		
	ng DW·cell ⁻¹	ng protein·cell ⁻¹	Ratio	ng DW·cell ⁻¹	ng protein·cell ⁻¹	Ratio	ng DW·cell ⁻¹	ng protein·cell ⁻¹	Ratio
0.003	2.166 $\pm$ 0.218	0.152 $\pm$ 0.050	0.070	1.172 $\pm$ 0.095	0.121 $\pm$ 0.008	0.103			
0.005	1.625 $\pm$ 0.246	0.149 $\pm$ 0.014	0.092	1.150 $\pm$ 0.087	0.130 $\pm$ 0.015	0.113	0.961 $\pm$ 0.206	0.117 $\pm$ 0.021	0.121
0.010	1.990 $\pm$ 0.151	0.138 $\pm$ 0.081	0.070	1.168 $\pm$ 0.070	0.175 $\pm$ 0.013	0.150	0.766 $\pm$ 0.052	0.134 $\pm$ 0.016	0.175
0.015	2.304 $\pm$ 0.125	0.182 $\pm$ 0.124	0.079	1.419 $\pm$ 0.135	0.186 $\pm$ 0.015	0.131	0.963 $\pm$ 0.062	0.139 $\pm$ 0.008	0.144
0.020	2.152 $\pm$ 0.208	0.310 $\pm$ 0.020	0.144	1.437 $\pm$ 0.144	0.196 $\pm$ 0.023	0.137	1.006 $\pm$ 0.075	0.145 $\pm$ 0.002	0.144
0.030				1.663 $\pm$ 0.298	0.222 $\pm$ 0.005	0.134	1.397 $\pm$ 0.121	0.230 $\pm$ 0.035	0.165
0.035							1.427 $\pm$ 0.177	0.242 $\pm$ 0.068	0.170

vantage to the oligotrophic species is likely to be compensated for by superiority in another physiological characteristic—for instance, the uptake of scarce nutrients. As phosphate (P) is often the limiting nutrient for algal growth in freshwaters during the summer period (e.g. Kalff and Knoechel 1978), this nutrient was chosen to test this hypothesis.

In the present study, three planktonic desmid species were grown under phosphorus-limited conditions. One of these taxa, *Cosmarium abbreviatum* var. *planctonicum*, is characteristic of an oligo-mesotrophic environment, whereas *Staurastrum pingue* and *Staurastrum chaetoceras* peak in eutrophic water bodies (Coesel 1994). These species were chosen as experimental subjects because of their roughly comparable cell dimensions, since cell size is an important parameter determining P uptake kinetics (Smith and Kalff 1982). Possible differences in kinetic characteristics, caused by cellular surface area-to-volume ratios are thus reduced to a minimum. We (Spijkerman and Coesel 1996) have shown that under stringent P limitation *C. abbreviatum* had a higher affinity for P uptake and growth than *S. pingue*. The maximum P uptake rates of *S. pingue* were higher than those of *C. abbreviatum* at all growth rates tested. For *C. abbreviatum* the total range of growth rates was examined, but for *S. pingue* only its kinetic characteristics up to about half its maximum growth rate ($\mu_{\max}$) were determined. Therefore, an extended set of kinetic data is provided here, including those for *S. chaetoceras*.

The species-specific uptake and growth kinetics from the unialgal P-limited chemostats were used to predict the outcome of competition. This was tested in mixed continuous flow cultures under both continuously and pulsed P-limited conditions. As far as we can determine, this is the first study of algal growth kinetics under P limitation comparing species with comparable cell size and belonging to the same taxonomic family but occurring under trophically different conditions in the field.

MATERIALS AND METHODS

The experiments were performed with *Staurastrum chaetoceras* (Schr.) G. M. Smith, clone AO 36, and *Staurastrum pingue* Teil., clone AO 234, both isolated from the alkaline eutrophic Lake IJmeer (Berger and Sweers 1988) and *Cosmarium abbreviatum* Rac.

var. *planctonicum* W. & G. S. West, clone AO 116, isolated from the alkaline, oligo-mesotrophic Lake Maarsseveen (I) (Swain et al. 1987). All clones were taken from the desmid collection at the Department of Aquatic Ecology.

Culture conditions and analytical methods. The species were cultured at 20° $\pm$ 1° C in 1-L continuous-flow culture vessels. The inflow medium for continuously P-limited cultures contained 5 μ M P; the inflow medium for the pulsed P-limited cultures contained no phosphorus. For details about the continuous-flow device, see Coesel and Wardenaar (1994). For details about the composition of the medium, see Spijkerman and Coesel (1996). The range of dilution rates (D) tested varied from 0.003 to 0.035 h⁻¹ for both *Staurastrum* species and from 0.003 to 0.020 h⁻¹ for *Cosmarium abbreviatum*. Circular fluorescent tubes provided an average photosynthetically active radiation in the culture vessel of 60–80 μ mol·m⁻²·s⁻¹, which proved to be saturating for growth. Illumination of the unialgal cultures was continuous. Cultures were not axenic, but bacterial biomass (estimated by acridine staining and counting under a fluorescence microscope) was negligible (<1% of algal biomass). Algal cells were counted with a Coulter Multisizer. Chemostat cultures were considered to be in a steady state (D = μ) when algal biomass varied less than 5% over at least two (in case of the lowest dilution rate) to five renewals of the culture vessel volume.

At steady-state conditions, protein was measured according to Lowry et al. (1951). Dry weight was measured by concentrating culture material on preweighed GF/C filters, which were dried at 70° C for 48 h. Cellular P concentrations were determined in the pellet after centrifugation (1500 $\times$ g, 10 min); total cellular P was measured after digestion with K₂S₂O₈ and H₂SO₄ at 100° C for 1 h. The external soluble reactive phosphorus (SRP) was assessed from the supernatant after centrifugation (1500 $\times$ g, 10 min). Both fractions were measured according to Murphy and Riley (1962). By dividing total cellular P concentration by cell density, cellular P quota (Q_p) were obtained. Tests for significance were done following Sokal and Rohlf (1981). In the data, no homogeneity in variance was obtained so comparing group means was done with a Kruskal and Wallis test and correlations were conducted with a Spearman test. Mean cellular dry weights and protein contents over the whole range of growth rates measured are given in Table 1 to enable conversion of the data.

Uptake experiments. Uptake experiments were performed with culture material from steady-state conditions, which was twofold diluted with P-free culture medium and pulsed with different inorganic phosphorus (P_i) concentrations containing ³²P as described in Spijkerman and Coesel (1996). Initial P_i concentrations ranged from 0.5 to 30 μ M. Cellular ³²P contents were determined at t = 0, 30, 60, and 300 s, and initial uptake rates (V) for every initial P_i concentration were calculated from linear regression, following Riegman and Mur (1984). By curve fitting to the Michaelis-Menten equation

$$V = V_{\max} \frac{P_i}{(K_m + P_i)}, \quad (1)$$

maximum uptake rate ($V_{\max}$) and half-saturation constant for uptake (K_m) were computed.

Calculation of chemostat culture parameters. In the cultures growing at low dilution rates, SRP concentrations in the culture vessel were too low to be measured accurately with the method of Murphy and Riley (1962); therefore, the external inorganic P concentrations were calculated from steady-state cellular quota and initial uptake rates, following Turpin (1988). In steady state, the net uptake rate of the limiting nutrient equals the dilution of cell quota due to growth:

$$V = (\mu)(Q_p). \quad (2)$$

From equation (1), the relationship between P_i and V had been obtained for a steady-state situation; thus, the actual P_i concentration for this steady-state condition in the culture vessel may be derived by combining equations (1) and (2), assuming that gross and net P_i influx are equal.

These calculated P_i values were used for the determination of the growth rate versus external P_i concentration expressed as a modified Monod model:

$$\mu = \mu_{\max} \frac{(P_i - P_i)}{(K_s + (P_i - P_i))}, \quad (3)$$

where P_i is the concentration of P_i at $\mu = 0$ (Tilman and Kilham 1976) and K_s is the half-saturation constant for growth.

The cellular P quotas were related to the growth rate according to the Droop relation:

$$\mu = \mu'_{\max} \left(1 - \frac{Q_0}{Q_p} \right), \quad (4)$$

where $\mu'_{\max}$ is the apparent maximal growth rate that would occur if Q_p became infinite and Q_0 is the cellular quota at $\mu = 0$.

Competition experiments. Competition between one *Staurastrum* species and *C. abbreviatum* was followed under both continuous and pulsed P limitation. To resemble natural conditions more closely, illumination in the competition experiments was applied under a 16:8 h LD regime. Other growth conditions were as described for single-species chemostat cultures. Algal strains were precultured in chemostats under the desired P limitation. Experiments were then started by mixing two species in a 1-to-1 cell number ratio. Cell numbers were counted one to three times a week using a 1-mL capacity Sedgewick-Rafter cell. Cell densities were recorded until one species dominated the culture for more than 95% of the total biomass.

Competition experiments between *S. pingue* and *C. abbreviatum* under continuous P limitation were performed at three different dilution rates, that is, 0.003, 0.007, and 0.020 h^{-1} . Competition between *C. abbreviatum* and *S. chaetoceras* under continuous P limitation was followed at $D = 0.007 \text{ h}^{-1}$ and $D = 0.017 \text{ h}^{-1}$.

Two discontinuously P-limited competition experiments with *C. abbreviatum* and *S. chaetoceras* were also carried out at $D = 0.007 \text{ h}^{-1}$. In one of these experiments, phosphate was added in a pulse of $2.5 \mu\text{mol P} \cdot \text{L}^{-1}$ of culture suspension two times a week; in the other, $0.7 \mu\text{mol P} \cdot \text{L}^{-1}$ of culture suspension was added every day. Pulses were always supplied in the light period. Both pulse regimes effected a total weekly P supply comparable to that in the continuously P-limited competition experiment at that same dilution rate. After 55 days, the condition in the culture growing at a small pulse regime was changed to a continuous P limitation.

RESULTS

Cellular quota. The cellular phosphorus quotas for the three species related to the growth rate (Fig. 1) were fitted to the Droop relation (equation (3)). The model estimated Q_0 values of 0.027, 0.030, and 0.026 $\text{pmol P} \cdot \text{cell}^{-1}$ for *C. abbreviatum*, *S. pingue*, and *S. chaetoceras*, respectively. There was no significant dif-

FIG. 1. Cellular P quota (Q_p) in relation to growth rate (μ) of A) *Cosmarium abbreviatum*, B) *Staurastrum pingue*, and C) *Staurastrum chaetoceras*.

ference in growth rate-related cellular P content at growth rates from 0.003 to 0.020 h^{-1} between the species ($P = 0.81$). Part of the variance in the data might be explained by the variance in cell volumes of the species. These volumes ranged for *S. chaetoceras* from 1500 to 4500 μm^3 , for *S. pingue* from 2000 to 4000 μm^3 , and for *C. abbreviatum* from 2500 to 5500 μm^3 . All species exhibited the smallest cell size at the lower growth rates.

FIG. 2. Growth rates (μ) in relation to the inorganic phosphorus concentration in the culture vessel (P_i) of A) *Cosmarium abbreviatum*, B) *Staurastrum pingue*, and C) *Staurastrum chaetoceras*. Curves fitted to the Monod model.

measurements in the culture vessel at $\mu = 0.007 \text{ h}^{-1}$, no significant difference in possible leakage of P among the three species was observed (data not shown).

Competition. During the continuous P limitation competition experiments between *S. pingue* and *C. abbreviatum*, there was a gradual replacement of *Staurastrum* by *Cosmarium* at the dilution rates 0.003

FIG. 3. Monod curves of the three desmid species as in Figure 2 to demonstrate the intersection at $P_i = 0.02 \text{ } \mu\text{M P}$.

and 0.007 h^{-1} (Fig. 4A, B). At $D = 0.020 \text{ h}^{-1}$, *S. pingue* replaced *C. abbreviatum* within a 50-day period (Fig. 4C). From these competition experiments, growth rates of *C. abbreviatum* were related to those of *S. pingue* resulting in a value greater than 1 at $D = 0.003$ and 0.007 h^{-1} and less than 1 at $D = 0.020 \text{ h}^{-1}$ (Fig. 5). The μ of *C. abbreviatum* relative to the μ of *S. pingue* at $D = 0.003 \text{ h}^{-1}$ was significantly higher than this relative growth rate at $D = 0.007 \text{ h}^{-1}$ ($P < 0.005$).

At $D = 0.007 \text{ h}^{-1}$, a competition experiment under a continuous P limitation between *C. abbreviatum* and *S. chaetoceras* resulted in the dominance of *C. abbreviatum* (Fig. 6). From this experiment (one of our preliminary tests), only relative cell numbers were gathered, but just as in the competition experiment between *S. pingue* and *C. abbreviatum* at that same dilution rate (Fig. 4B) *S. chaetoceras* was replaced by *Cosmarium* within 80 days. At $D = 0.017 \text{ h}^{-1}$, in contrast, competition between these same two species resulted in a marked predominance of *S. chaetoceras* within 40 days (Fig. 7). The mean growth rate of *C. abbreviatum* relative to that of *S. chaetoceras* in this experiment was 0.84.

In the pulsed P competition experiment at $D = 0.007 \text{ h}^{-1}$, at a relatively big pulse two times a week, *S. chaetoceras* displaced *C. abbreviatum* over an 80-day period (Fig. 8). The mean relative growth rate of *C. abbreviatum* compared to *S. chaetoceras* over this period was 0.84. Also in the experiment with a relatively small pulse given every day, *S. chaetoceras* soon acquired a dominant position (Fig. 9). However, after 2 weeks a condition was attained in which abundances of *S. chaetoceras* and *C. abbreviatum* hardly changed any more. The mean growth rate of *C. abbreviatum* relative to that of *S. chaetoceras* over the period from day 13 to day 55 was 0.96, implying

FIG. 4. Cell concentrations in mixed continuous flow cultures of *Cosmarium abbreviatum* (O) and *Staurastrum pingue* (●) in relation to time at different dilution rates (D). A) $D = 0.003 \text{ h}^{-1}$, B) $D = 0.007 \text{ h}^{-1}$, and C) $D = 0.020 \text{ h}^{-1}$. Slopes ($\mu = 0$) indicate expected decrease rates over a period of 10 days for nongrowing organisms.

almost stable coexistence. After day 55, the culture was brought into a continuous P limitation and within 40 days *C. abbreviatum* predominated the culture in cell number for 85%.

DISCUSSION

When P uptake and growth kinetics of the three desmid taxa were compared, a striking result was

FIG. 5. Growth rate of *Cosmarium abbreviatum* relative to that of *Staurastrum pingue* measured in mixed continuous-flow cultures at three different dilution rates.

the high degree of similarity between *Staurastrum pingue* and *Staurastrum chaetoceras*, both distinctly deviating in their kinetics from *Cosmarium abbreviatum*. All three Monod relations estimate maximum specific growth rates comparable to the previously determined rates of 0.051, 0.048, and 0.020 h^{-1} for *S. chaetoceras*, *S. pingue*, and *C. abbreviatum*, respectively (Coesel and Wardenaar 1990). Under conditions of stringent, continuous P limitation, the affinity to P for growth was significantly higher in *Cosmarium* than in *Staurastrum*. Under a given condition of stringent continuous P limitation at which *Cosmarium* has lowered the external P concentration

FIG. 6. Ratio in cell number between *Cosmarium abbreviatum* (O) and *Staurastrum chaetoceras* (■) in relation to time in a mixed continuous-flow culture at $D = 0.007 \text{ h}^{-1}$.

FIG. 7. Cell concentrations in mixed continuous-flow cultures of *Cosmarium abbreviatum* (○) and *Staurastrum chaetoceras* (■) in relation to time at $D = 0.017 \text{ h}^{-1}$. Slope ($\mu = 0$) indicates expected decrease rate over a period of 10 days for nongrowing organisms.

to a certain value, *Cosmarium* can realize a much higher growth rate relative to those of both *Staurastrum* species. *Cosmarium* is thus better adapted to growth in situations with continuously low P concentrations as defined for oligo-mesotrophic lakes. Both *Staurastrum* species will be outcompeted by *Cosmarium* under oligotrophic conditions.

This characteristic to acquire P at low P concentrations is caused by the higher affinity to P for uptake of *C. abbreviatum* compared to both *Staur-*

FIG. 8. Cell concentrations in mixed continuous-flow culture of *Cosmarium abbreviatum* (○) and *Staurastrum chaetoceras* (■) in relation to time at $D = 0.007 \text{ h}^{-1}$, under a pulsed P addition of $2.5 \mu\text{mol P}$ per liter of culture two times a week. Slope ($\mu = 0$) indicates expected decrease rate over a period of 10 days for nongrowing organisms.

FIG. 9. Cell concentrations in mixed continuous-flow culture of *Cosmarium abbreviatum* (○) and *Staurastrum chaetoceras* (■) in relation to time at $D = 0.007 \text{ h}^{-1}$ under different P-limited conditions. I) Pulsed P addition of $0.7 \mu\text{mol P}$ per liter of culture every day; II) continuous P limitation.

astrum species. Under stringent P limitation, the cell-based P quotas of the three species were comparable. Because Q_0 represents the amount of P associated with the structural and metabolic components that are essential to cellular integrity and viability (e.g. cell walls and membranes, DNA, machinery responsible for maintenance metabolism) and the absolute amounts of P increase with increasing cell size (Smith and Kalff 1982), a size dependence for Q_0 can be expected (Fuhs 1969, Droop 1974, Goldman 1977). As already stated, cell size of the species studied was about equal. Dry weight-based yields, however, indicate a difference among the species. A difference in mean cellular weight of 2.0, 1.3, and $1.0 \text{ ng DW} \cdot \text{cell}^{-1}$ for *C. abbreviatum*, *S. pingue*, and *S. chaetoceras*, respectively, could be calculated. *Cosmarium abbreviatum* has a copious mucilage sheath around the cell, whereas *S. pingue* and *S. chaetoceras* do not have such a sheath (Coesel 1994). Because such an extracellular mucus envelope may contain a substantial amount of carbohydrates (e.g. Surek 1983, Paulsen and Viera 1994), *C. abbreviatum* is likely to produce a higher biomass weight per unit of P than the two *Staurastrum* species. Even the slightly higher biomass yield of *S. pingue* as compared to *S. chaetoceras* might be related to this feature because cells of *S. pingue* showed some mucilage layer whereas cells of *S. chaetoceras* completely lacked it. The cellular dry weight may thus (partly) reflect the amount of mucus surrounding the cell, which makes *C. abbreviatum* cells substantially heavier than the cells of both *Staurastrum* species. The influence of carbohydrate production on cellular weight was also noticed by Kromkamp et al. (1989). Their study showed that *Microcystis aeruginosa* produced a higher Q_p (based

on dry weight) at low dilution rates, when compared to Q_p expressed per unit of protein. Dry weight may thus suggest more biomass production even though this is not translated into more protein or cells.

The observed species-specific differences in P growth and uptake kinetics fit very well with the origin of the algal strains used because the two *Staurastrum* species were isolated from an alkaline, eutrophic lake and *Cosmarium abbreviatum* from a likewise alkaline but oligo-mesotrophic lake. Their P kinetics reflect the "ghost of competition past" (Connell 1980, Sommer 1989) when considering P limitation an important factor determining algal dominance in an oligo-mesotrophic lake. The result of the competition experiments between *Cosmarium abbreviatum* and *Staurastrum pingue* or *Staurastrum chaetoceras* can be explained from the species-specific Monod relations. The outcome of our competition experiments agrees with the prediction that the species that can deplete the medium to the lowest concentration should displace all competitors (O'Brien 1974, Tilman 1977). The Monod relation can be interpreted as defined by Tilman et al. (1981), where P_i is replaced by the equivalent symbol R^* (residual nutrient concentration) and, thus, the species with the lowest R^* for the limiting nutrient wins under continuously P-limited conditions. Where the species with the lowest R^* value determines the external P_i concentration, it also determines indirectly the growth rates possible for all other competitors. From Figure 3 it can be derived that $D = 0.003 \text{ h}^{-1}$ will result in a P_i of $0.003 \mu\text{M}$ where hardly any growth of *S. pingue* or *S. chaetoceras* will be possible, whereas at $D = 0.007 \text{ h}^{-1}$, resulting in a P_i concentration of $0.007 \mu\text{M}$, *Staurastrum* would be able to grow at a relative rate of 0.6 compared to *Cosmarium*. The competitive advantage of *C. abbreviatum* compared to *S. pingue* would thus be greater at $D = 0.003 \text{ h}^{-1}$ than at $D = 0.007 \text{ h}^{-1}$, which is confirmed by Figure 5.

Competition experiments between *S. chaetoceras* and *C. abbreviatum* under continuous P limitation at $D = 0.007 \text{ h}^{-1}$ and $D = 0.017 \text{ h}^{-1}$ showed outcomes comparable to those between *S. pingue* and *Cosmarium* at these dilution rates. Mean growth rates of *C. abbreviatum* at $D = 0.007 \text{ h}^{-1}$ were 1.35 and 1.41 relative to those of *S. chaetoceras* and *S. pingue*, respectively, and, at $D = 0.017\text{--}0.020 \text{ h}^{-1}$, 0.84 relative to both *S. chaetoceras* and *S. pingue*. These results confirm that there were no significant differences between the two *Staurastrum* species in these competition experiments as could be predicted from their Monod relations. Although it is theoretically possible that the Monod relations, as determined in monospecies cultures under continuous illumination are somewhat altered under the light-dark cycle as imposed in the competition experiments (see e.g. Chisholm and Stross 1976a, b), we have no indication of it. P uptake kinetics and P cell quota of suspensions from some competition experiments con-

sisting of 95% of one species were comparable to those from monospecies experiments under continuous light (data not shown). The fact that the outcome of competition in our mixed chemostat experiments nicely matched with what was expected from the various species-specific affinities, as determined in single-species cultures, makes any species interference (allelopathic interactions) unlikely. Neither was there any indication of possible differences in cellular P leakage between the two *Staurastrum* species as observed from the exclusion rates in the various competition experiments.

Literature on competition experiments between algal species related to their nutrient uptake and growth kinetics is scarce and as far as desmids are concerned only the studies of Healey and Hendzel (1988) and Olsen et al. (1989) can be mentioned. Healey and Hendzel (1988) compared *Cosmarium subcostatum* and *Staurastrum paradoxum* (originating from the same lake) with regard to uptake and growth kinetics under P limitation. *Cosmarium subcostatum* appeared to dominate at $D = 0.008 \text{ h}^{-1}$, which was ascribed to its greater yield of biomass per unit of P rather than to a higher affinity to P for uptake. Presumably, coexistence of *S. paradoxum* and *C. subcostatum* within one and the same lake was enabled by differences in other responses (e.g. vulnerability to predation). Cell volume of *S. paradoxum* was calculated to be two to three times that of *C. subcostatum*. The competitive advantage in resource competition of the smaller sized *C. subcostatum* over the larger sized *S. paradoxum*, as demonstrated by Healey and Hendzel (1988), agrees with the conclusion by Smith and Kalff (1983) that competition at a wide range of continuous nutrient limitations always favors the smaller alga because superiority in $\mu_{\max}$ would go hand in hand with a superiority in competitive ability. The untenability of this conclusion was argued by, for example, Sommer and Kilham (1985) and also demonstrated in competition experiments with natural plankton by Sommer (1985). Cell size may be a major factor in predicting competition between phytoplankton species for a limiting nutrient, but the importance of variation in uptake parameters among species of comparable size should not be underestimated. Our study unequivocally shows that also within a taxonomically uniform algal group the species with the highest $\mu_{\max}$ will not per se be superior in competition.

The competition experiments by Olsen et al. (1989) showed that *Staurastrum luetkemulleri* with a higher $\mu_{\max}$ and a higher affinity to P over the whole range of growth rates wins competition under a continuous P limitation but loses to the blue-green *Microcystis aeruginosa* under markedly pulsed conditions due to the 10 times lower $V_{\max}$ of *Staurastrum*. The prediction that the species with the higher $V_{\max}$ wins competition under pulsed conditions (Eppley et al. 1969, Turpin and Harrison 1979, Suttle et al. 1987) is also confirmed in our experiment where the spe-

cies was given a pulse of $2.5 \mu\text{mol P} \cdot \text{L}^{-1}$ twice a week. In our experiment where a daily pulse of $0.7 \mu\text{mol P} \cdot \text{L}^{-1}$ was given, a stable coexistence of the species was established during the last 40 days of the experiment.

Stewart and Levin (1973) theoretically demonstrated that coexistence of species competing for the same resource can occur under pulsed conditions. In addition, Tilman (1982) reasoned that fluctuations in absolute resource level can enable coexistence and thus enhance species diversity. The latter was nicely illustrated by Sommer (1985) with natural phytoplankton assemblages from Lake Constance. Our data indicate that also between the *Cosmarium* and *Staurastrum* clones used in our experiments a dynamic equilibrium can be found under a specific, carefully balanced pulse regime.

- Berger, C. & Sweers, H. E. 1988. The IJsselmeer and its phytoplankton—with special attention to the suitability of the lake as a habitat for *Oscillatoria agardhii* Gom. *J. Plankton Res.* 10:579–99.
- Chisholm, S. W. & Stross, R. G. 1976a. Phosphate uptake kinetics in *Euglena gracilis* (Z) (Euglenophyceae) grown on light/dark cycles. I. Synchronized batch cultures. *J. Phycol.* 12:210–7.
- 1976b. Phosphate uptake kinetics in *Euglena gracilis* (Z) (Euglenophyceae) grown in light/dark cycles. II. Phased PO_4 -limited cultures. *J. Phycol.* 12:217–22.
- Coesel, P. F. M. 1994. On the ecological significance of a cellular mucilaginous envelope in planktic desmids. *Algological Studies* 73:65–74.
- Coesel, P. F. M. & Wardenaar, K. 1990. Growth responses of planktonic desmid species in a temperature-light gradient. *Freshwater Biol.* 23:551–60.
- 1994. Light-limited growth and photosynthetic characteristics of two planktonic desmid species. *Freshwater Biol.* 31:221–6.
- Connell, J. H. 1980. Diversity and the coevolution of competitors, or the ghost of competition past. *Oikos* 35:131–8.
- Droop, M. R. 1974. The nutrient status of algal cells in continuous culture. *J. Mar. Biol. Assoc. U.K.* 54:825–55.
- Eppley, R. W., Rogers, J. N. & McCarthy, J. J. 1969. Half-saturation constants for uptake of nitrate by marine phytoplankton. *Limnol. Oceanogr.* 14:912–20.
- Fuhs, G. W. 1969. Phosphorus content and rate of growth in the diatoms *Cyclotella nana* and *Thalassiosira fluciatilis*. *J. Phycol.* 5:312–21.
- Goldman, J. C. 1977. Steady state growth of phytoplankton in continuous culture: comparison of internal and external nutrient equations. *J. Phycol.* 13:251–8.
- Grover, J. P. 1989a. Phosphorus-dependent growth kinetics of 11 species of freshwater algae. *Limnol. Oceanogr.* 34:341–8.
- 1989b. Effects of Si:P supply ratio, supply variability, and selective grazing in the plankton: an experiment with a natural algal and protistan assemblage. *Limnol. Oceanogr.* 34:349–67.
- 1991. Algae grown in non-steady continuous culture: population dynamics and phosphorus uptake. *Verh. Int. Verein. Limnol.* 24:2661–4.
- Healey, F. P. & Hendzel, L. L. 1988. Competition for phosphorus between desmids. *J. Phycol.* 24:287–92.
- Hecky, R. E. & Kilham, P. 1974. Environmental control of phytoplankton cell size. *Limnol. Oceanogr.* 19:361–6.
- Holm, N. P. & Armstrong, D. E. 1981. Role of nutrient limitation and competition in controlling the populations of *Asterionella formosa* and *Microcystis aeruginosa* in semicontinuous culture. *Limnol. Oceanogr.* 26:622–34.
- Kalff, J. & Knoechel, R. 1978. Phytoplankton and their dynamics in oligotrophic and eutrophic lakes. *Annu. Rev. Ecol. Syst.* 9:475–95.
- Kilham, P. & Tilman, D. 1979. The importance of resource competition and nutrient gradients for phytoplankton ecology. *Ergeb. Limnol.* 13:110–9.
- Kilham, S. S. 1984. Silicon and phosphorus growth kinetics and competitive interactions between *Stephanodiscus minutus* and *Synedra* sp. *Verh. Int. Verein. Limnol.* 22:435–9.
- Kromkamp, J., van den Heuvel, A. & Mur, L. R. 1989. Phosphorus uptake and photosynthesis by phosphate-limited cultures of the cyanobacterium *Microcystis aeruginosa*. *Br. Phycol. J.* 24:347–55.
- Lowry, O. H., Rosebrough, N. J., Farr, A. L. & Randall, R. J. 1951. Protein measurement with the folin phenol reagent. *J. Biol. Chem.* 193:265–75.
- Murphy, J. & Riley, J. P. 1962. A modified single-solution method for the determination of phosphate in natural waters. *Anal. Chim. Acta* 27:31–6.
- Nicklisch, A., Roloff, B. & Ratsch, A. 1991. Competition experiments with two planktic blue-green algae (Oscillatoriaceae). *Verh. Int. Verein. Limnol.* 24:889–92.
- O'Brien, W. J. 1974. The dynamics of nutrient limitation of phytoplankton algae: a model reconsidered. *Ecology* 55:135–41.
- Olsen, Y. 1989. Evaluation of competitive ability of *Staurastrum luetkemullerii* (Chlorophyceae) and *Microcystis aeruginosa* (Cyanophyceae) under P limitation. *J. Phycol.* 25:486–99.
- Olsen, Y., Vadstein, O., Andersen, T. & Jensen, A. 1989. Competition between *Staurastrum luetkemullerii* (Chlorophyceae) and *Microcystis aeruginosa* (Cyanophyceae) under varying modes of phosphate supply. *J. Phycol.* 25:499–508.
- Paulsen, B. S. & Viera, A. A. H. 1994. Structure of the capsular and extracellular polysaccharides produced by the desmid *Spondylium panduriforme* (Chlorophyta). *J. Phycol.* 30:638–41.
- Riegman, R. & Mur, L. R. 1984. Regulation of phosphate uptake kinetics in *Oscillatoria agardhii*. *Arch. Microbiol.* 139:28–32.
- Smith, R. E. H. & Kalff, J. 1982. Size-dependent phosphorous uptake kinetics and cell quota in phytoplankton. *J. Phycol.* 18:275–84.
- 1983. Competition for phosphorus among co-occurring freshwater phytoplankton. *Limnol. Oceanogr.* 28:448–64.
- Sokal, R. R. & Rohlf, F. J. 1981. *Biometry*, 2nd ed. W. H. Freeman, New York, 859 pp.
- Sommer, U. 1985. Comparison between steady state and non-steady state competition: experiments with natural phytoplankton. *Limnol. Oceanogr.* 30:335–46.
- 1989. The role of competition for resources in phytoplankton succession. In Sommer, U. [Ed.] *Plankton Ecology: Succession in Plankton Communities*. Springer-Verlag, New York, pp. 57–107.
- Sommer, U. & Kilham, S. S. 1985. Phytoplankton natural community experiments. A reinterpretation. *Limnol. Oceanogr.* 30:436–40.
- Spijkerman, E. & Coesel, P. F. M. 1996. Phosphorus uptake and growth kinetics of two planktonic desmid species. *Eur. J. Phycol.* 31:53–60.
- Stewart, F. M. & Levin, B. R. 1973. Partitioning of resources and the outcome of interspecific competition: a model and some general considerations. *Am. Nat.* 107:171–98.
- Surek, B. 1983. Mucilage regeneration in the green alga *Cosmoecium saxonicum* De Bary (Desmidiaceae); a light microscopic and quantitative study. *Br. Phycol. J.* 18:73–81.
- Suttle, C. A., Stockner, J. G. & Harrison, P. J. 1987. Effects of nutrient pulses on community structure and cell size of a freshwater phytoplankton assemblage in culture. *Can. J. Fish. Aquat. Sci.* 44:1768–74.
- Swain, W. R., Lingeman, R. & Heinis, F. 1987. A characterization and description of the Maarsseveen Lake system. *Hydrobiol. Bull.* 21:5–16.
- Tilman, D. 1977. Resource competition between planktonic algae: an experimental and theoretical approach. *Ecology* 58:338–48.

- . 1981. Tests of resource competition theory using four species of Lake Michigan algae. *Ecology* 62:802–15.
- . 1982. *Resource Competition and Community Structure*. Princeton University Press, Princeton, New Jersey, 296 pp.
- Tilman, D. & Kilham, S. S. 1976. Phosphate and silicate growth and uptake kinetics of the diatoms *Asterionella formosa* and *Cyclotella meneghiniana* in batch and semicontinuous culture. *J. Phycol.* 12:375–83.
- Tilman, D., Kilham, S. S. & Kilham, P. 1982. Phytoplankton community ecology: the role of limiting nutrients. *Ann. Rev. Ecol. Syst.* 13:349–72.
- Tilman, D. Mattson, M. & Langer, S. 1981. Competition and nutrient kinetics along a temperature gradient: an experimental test of a mechanistic approach to niche theory. *Limnol. Oceanogr.* 26:1020–33.
- Turpin, D. H. 1988. Physiological mechanisms in phytoplankton resource competition. In Sandgren, C. D. [Ed.] *Growth and Reproductive Strategies of Freshwater Phytoplankton*. Cambridge University Press, Cambridge, pp. 316–68.
- Turpin, D. H. & Harrison, P. J. 1979. Limiting nutrient patchiness and its role in phytoplankton ecology. *J. Exp. Mar. Biol. Ecol.* 39:151–66.

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