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Silicon and the Ecology of Marine Plankton Diatoms. II. Silicate-Uptake Kinetics in Five Diatom Species

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Abstract

The variation of the rate of silicate uptake with varying silicate concentration in the medium was investigated in short-term experiments with the following marine diatom species: *Skeletonema costatum, Thalassiosira pseudonana, T. decipiens, Ditylum brightwellii*, and *Licmophora* sp. The uptake conformed to Michaelis-Menten kinetics only after a correction had been made for reactive silicate that apparently could not be utilized by the diatoms. The magnitude of this correction was in the range of 0.3 to 1.3 µg-at Si/l. Mean values of the half-saturation constant of silicate uptake were calculated for the different species. The lowest value was found in *S. costatum* (0.80 µg-at Si/l) and the highest in *T. decipiens* (3.37 µg-at Si/l). Growth limitation by low silicate concentrations could be a cause of species succession in marine plankton-diatom blooms.

Introduction

In the first part of this investigation (Paasche, 1973), chemostats were used to establish the relationship between the growth rate of a marine plankton diatom, *Thalassiosira pseudonana*, and the content of dissolved silicate in the medium. Apart from this paper and a growth study by Guillard *et al.* (in press) using the same species, there is little information in the literature on the silicate requirements of marine plankton diatoms.

In recent years, short-term uptake experiments with nitrate and ammonia have been successfully used to demonstrate differences between species (Eppley *et al.*, 1969), or even between strains of one and the same species (Carpenter and Guillard, 1971), with regard to their uptake kinetics. The methods developed in these studies were adapted for the present purpose of comparing the silicate-uptake kinetics of 5 marine diatom species.

Materials and Methods

Bacteria-free cultures of *Thalassiosira pseudonana* Hasle and Heimdal (*Cyclotella nana* Hustedt) and of *Ditylum brightwellii* (West) Grunow were originally obtained from Dr. R. R. L. Guillard, Woods Hole Oceanographic Institution, and from Dr. T. J. Smayda, University of Rhode Island, respectively. Bacteria-free cultures of *Skeletonema costatum* (Greville) Cleve, *Thalassiosira decipiens* (Grunow) Jørgensen, and *Licmophora* sp. [probably *L. hyalina* (Kützing) Grunow] were isolated from the Oslo Fjord in June, 1970. These and other diatoms grew when a plankton concentrate was plated on the medium of Eppley *et al.* (1967), based on 70% sea water and solidified with agar. Bacteria-free clones were established by repeated streaking of diatom colonies picked from agar plates. Plankton diatoms usually are not expected to grow on solid media. Certain precautions were taken in the present work, and may have contributed to the success of the plating method: the agar was of good quality (Oxoid 'Ionagar No. 2') and was used at low concentrations (0.8%); it was autoclaved apart from the medium (agar and glass-distilled water in one flask, sea water and enrichments in another); drying-out of the agar was prevented by keeping the dishes in clear polyethylene bags during growth.

The cultures used in experiments were grown in 600 to 750 ml lots in "Polycarbonate" (Nalge Co.) Erlenmeyer flasks. The medium was that of Eppley *et al.* (1967), with the enrichments, less silicate, added at half strength. The medium was prepared from aged and membrane-filtered Oslo Fjord water diluted to 24‰ S with glass-distilled water. It was autoclaved for 1 min at 120 °C, with phosphate autoclaved separately, to reduce precipitation as much as possible. The reactive silicate content of the medium was ca. 5 µg-at Si/l.

The flasks were inoculated usually with 10 to 25 ml of a diatom culture grown in silicate-rich (ca. 500 µg-at Si/l) medium. All cultures were grown at 20 °C at a light intensity of about 25,000 erg/cm² · sec of white fluorescent light. An 18:6 h light-dark cycle was used with *Ditylum brightwellii*, which does not grow well in continuous light. The other species were illuminated continuously. Cell concentrations were measured by electronic particle counting or by means of a Palmer-Maloney counting chamber or a haemacytometer. Silicate in the medium was determined according to Strickland and Parsons (1968), and silica in the cells according to Werner (1966). Details of these procedures were described in the previous paper in the series (Paasche, 1973).

The uptake experiments were carried out as soon as the silicate content in the medium had been reduced from the 15 to 25 µg-at Si/l originally present (most of it being due to carry-over with the inoculum) to 0.5—1.5 µg-at Si/l. This occurred 2 to 3 days after inoculation. The culture was subdivided into 10 or 12 50 ml aliquots in 125 ml "Polycarbonate" flasks. To these was added from 0.05 to 1.5 ml of a 0.5 mM solution of $Na_2SiO_3 \cdot 9H_2O$ freshly neutralized with HCl. Two flasks received no addition, and one of these was deep-frozen and again thawed. This latter treatment caused no immediate change in the reactive silicate level (see Paasche, 1973).

Samples for reactive silicate analysis were immediately withdrawn from all the flasks. The flasks were then left for 1 h under the light and temperature conditions used for growth, and the medium was again analysed. In most experiments, the flasks were subjected to a second period of incubation followed by a final analysis of silicate in the medium. The silicate uptake rate per cell and hour was calculated from the decrease in the reactive silicate content of the medium. In the flask in which the cells had been inactivated by freezing, the uptake rate calculated in this way was always negative and was used as a correction for silica dissolution (see "Results"). According to Lewin (1961), any treatment that kills the cells also leads to an increased rate of dissolution of the silica cell-walls. Tests with all 5 species used in the present work showed that the rate of dissolution was usually smaller after the cultures had been frozen and thawed than after heating or treatment with chemicals.

Experience gained in the course of the work made it possible to predict fairly accurately the size of initial inoculum needed to produce a culture of suitable density on the day of the uptake experiment. In this way, conditions were arranged so that, in nearly all cases, between 5 and 20% of the silicate present in the medium was taken up during a 1 h incubation period.

The silicate-uptake rates were related to values expressing the silicate concentration in the middle of each incubation period. These latter values were calculated separately for each flask from the silicate concentration at the beginning and at the end of the incubation period, assuming a linear uptake.

The constants of the hyperbolic uptake curves were calculated by weighted regression analysis according to Wilkinson (1961).

Results

Figs. 1—3 show examples of how cell concentration, cell silica, and reactive silicate in the medium changed during growth. In all experiments of this kind, net silicate uptake ceased when there was still 0.5 to 1.5 µg-at Si/l left in the medium. Sometimes marked oscillations in the level of residual silicate could be detected by repeated analysis, even within as little as a few hours.

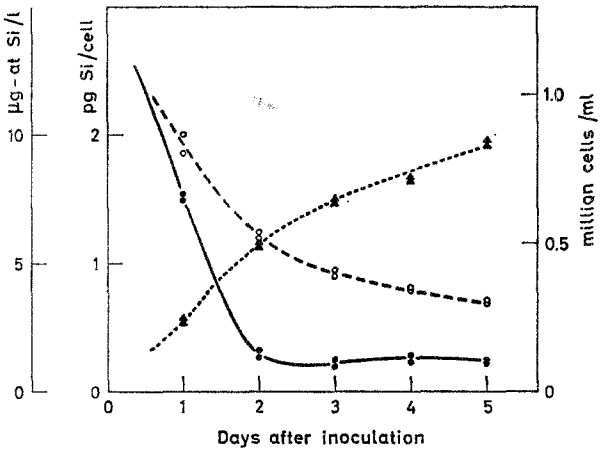

Fig. 1. *Skeletonema costatum.* Growth with limited silicate. Closed circles: silicate concentration in medium. Open circles: silica content/cell. Triangles: cell concentration

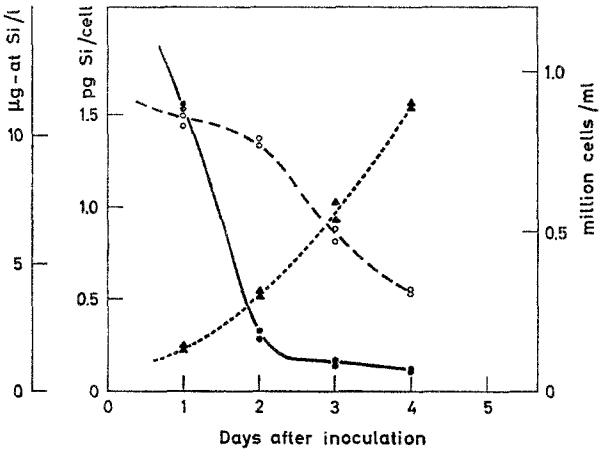

Fig. 2. *Thalassiosira pseudonana.* Growth with limited silicate. Symbols as in Fig. 1

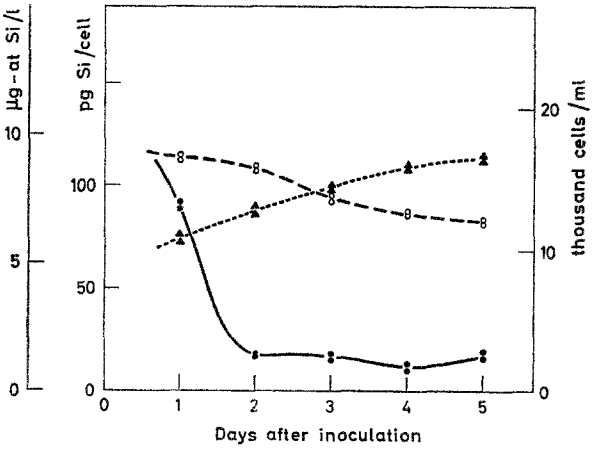

Fig. 3. *Thalassiosira decipiens.* Growth with limited silicate. Symbols as in Fig. 1

Especially in *Skeletonema costatum* (Fig. 1) and *Thalassiosira pseudonana* (Fig. 2), cell multiplication continued even after the cessation of net silicate uptake, although the growth rates were only about one-fifth of the maximum growth rates recorded with these species. In experiments with the fresh-water diatom *Navicula pelliculosa*, growth stopped abruptly when silicate became exhausted (Busby and Lewin, 1967). Vegetative cell division in diatoms normally cannot proceed in the absence of silicate uptake and valve formation. Most probably, the silica shells of *S. costatum* and *T. pseudonana* redissolved to some extent in the medium, whereby sufficient silicate became available to permit the formation of new valves. The following conditions would favour the process of dissolution and reutilization: the alkaline reaction of sea water; the thinness and delicate structure of the shells of these species (implying a large surface-to-volume ratio of the shell silica); a highly efficient silicate uptake mechanism; and a high degree of flexibility with regard to the amount of silica required to form a serviceable valve. Scanning electron microscopy of *S. costatum* cells harvested on the fifth day of growth indicated that newly formed cell walls were extremely weakly silicified, to the point where the valves were unable to retain their semi-spherical shape upon drying and the "rods" or tubuli connecting them did not develop properly. This latter phenomenon has been described previously from silicate-starved *S. costatum* cultures (Braarud, 1948).

The uptake experiments were carried out with silicate-depleted cultures. As a result, a net silicate uptake was usually observed only in flasks to which fresh silicate had been added. It was necessary, therefore, to apply a correction for silica dissolution in order to arrive at the true uptake rates. The rate of reappearance of reactive silicate in cultures inactivated by freezing and thawing was assumed to provide an estimate of the rate of silicate reutilization in living cultures. All uptake rates were corrected accordingly. The correction (denoted by a in Fig. 4) usually amounted to 10 to 20% of the maximum (saturated) silicate-uptake rates.

Even the corrected uptake rates, when plotted against the reactive silicate concentration, failed to produce simple saturation curves starting from zero silicate concentration. There was a choice of the following interpretations: (1) the uptake curves were sigmoidal, although asymmetric with an inflexion point near the abscissa; or (2) they were hyperbolic and of the form prescribed by Michaelis-Menten kinetics, although intersecting the abscissa at a positive silicate concentration. In this case, the curves should obey the mathematical expression:

$$v = \frac{V_{max} \cdot (Si - Si_0)}{K_v + (Si - Si_0)} \quad (1),$$

in which v is the uptake rate (as pg Si/cell · h); V_{max} the maximum or saturated uptake rate; K_v the half-saturation constant of uptake; Si the reactive silicate concentration; and Si_0 the silicate concentration when $v = 0$.

It turned out that, in most cases, a very good hyperbolic fit of this type could be obtained by the proper choice of a Si_0 value, suggesting that Michaelis-Menten kinetics provided a valid description of the kinetics of silicate uptake. Accordingly, the following procedure was adopted. The data from each experiment were plotted as $(Si - Si_0)/v$ versus $(Si - Si_0)$; this represents a linearized form of the Michaelis-Menten hyperbola particularly suited to data of the present type (see Eppley *et al.*, 1969). By a trial-and-error process, the correct Si_0 was selected from several likely values; it was identified as the value that caused all the points in the linearized plot to form as nearly a straight line as possible. The next step was to subtract this Si_0 from all the Si values and then to calculate statistically the constants, V_{max} and K_v. The diagram in Fig. 4 illustrates the interconnection of V_{max}, K_v, Si_0, and the dissolution correction, a.

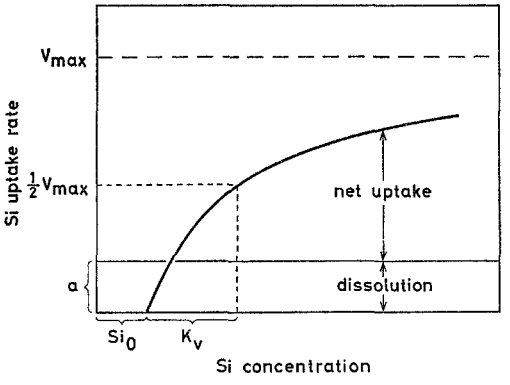

Fig. 4. Relationship of V_{max}, K_v, Si_0, and a. For further explanation, see text

The calculated constants from all the experiments are summarized in Table 1, and representative uptake curves are shown in Figs. 5—9. In *Ditylum brightwellii* and *Thalassiosira pseudonana*, an increase in K_v from the first to the second part of the experiment was noted in all cases, and was mostly associated with an increase in V_{max}. The main reason for this appeared to be a non-linear silicate uptake at high silicate concentrations. In fact, there was little reason to expect the uptake rate to remain absolutely constant throughout incubation. The exhaustion of silicate during growth and the readdition of silicate at the beginning of incubation resemble the conditions used by Lewin and her collaborators (Lewin *et al.*, 1966; Busby and Lewin, 1967) to induce "silicate starvation synchrony" in *Navicula pelliculosa*. The silicate-uptake curves in their experiments were distinctly non-linear. How-

Table 1. *Values of Si_0, K_v (± 95% confidence limits), and V_{max} of silicate uptake. Also included are data on cell size (diameter or length of valve) and cell silica content in Thalassiosira pseudonana (from Paasche, 1973) and in the 4 other species (from Paasche, in press). First and second half of an experiment are indicated by letters A and B*

Species	Cell size (µm)	Silica content (pg Si/cell)	Experiment No.	Si_0 (µg-at Si/l)	K_v (µg-at Si/l)	V_{max} (pg Si/cell · h)
Skeletonema costatum	4—6	3.8—7.0	1	0.20	0.80 ± 0.26	0.117
			2A	0.20	1.11 ± 0.44	0.092
			2B	0.20	0.42 ± 0.28	0.083
			3A	0.50	0.94 ± 0.20	0.100
			3B	0.50	0.71 ± 0.26	0.085
			Mean	0.32	0.80	0.095
Thalassiosira pseudonana	3—6	1.81	4A	0.40	1.00 ± 0.41	0.056
			4B	0.40	1.80 ± 0.39	0.084
			5A	0.50	1.07 ± 0.31	0.058
			5B	1.00	1.40 ± 0.55	0.040
			6A	0.90	0.91 ± 0.07	0.076
			6B	0.80	2.13 ± 0.42	0.127
			Mean	0.67	1.39	0.073
Licmophora sp.	14—40	80—210	7A	1.50	3.52 ± 1.06	4.20
			7B	1.50	2.06 ± 0.54	2.46
			8	1.00	1.53 ± 0.56	1.08
			9	1.20	3.19 ± 0.74	0.84
			Mean	1.30	2.58	2.15
Ditylum brightwellii	13—52	200—900	10A	0.50	2.59 ± 1.66	26.1
			10B	0.40	4.50 ± 1.21	37.7
			11A	0.50	2.35 ± 0.40	25.8
			11B	0.50	3.23 ± 0.79	27.6
			12A	0.10	1.85 ± 0.76	17.2
			12B	0.30	3.24 ± 0.68	24.9
			Mean	0.38	2.96	26.6
Thalassiosira decipiens	17—28	150—330	13A	1.20	2.97 ± 1.24	4.00
			13B	1.20	3.02 ± 1.13	4.96
			14A	1.00	5.65 ± 1.56	4.59
			14B	1.20	3.69 ± 1.30	4.33
			15A	1.60	2.52 ± 1.03	3.72
			15B	1.80	2.38 ± 1.19	2.93
			Mean	1.33	3.37	4.09

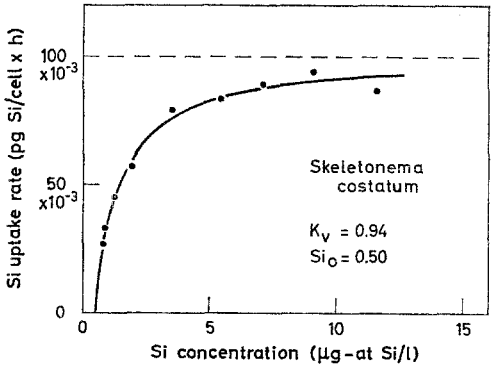

Fig. 5. *Skeletonema costatum.* Silicate-uptake rate as function of silicate concentration. Broken line: maximum rate

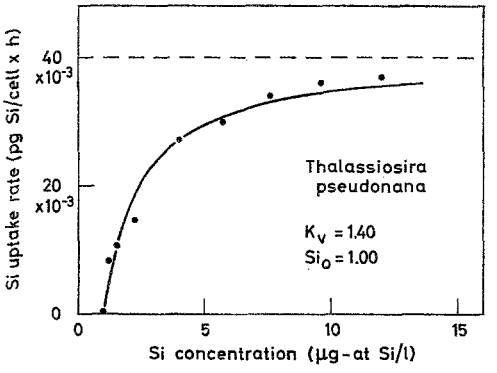

Fig. 6. *Thalassiosira pseudonana.* Silicate-uptake rate as function of silicate concentration. Broken line: maximum rate

ever, the present experiments differed from those of Lewin and associates in two respects. First, cell division was not brought to a complete halt by the exhaustion of silicate (Figs. 1—3). Second, the "starvation" period elapsing from the cessation of net silicate uptake to the readdition of silicate was made as short as possible. Perhaps as a consequence of this, no tendency towards synchronization could be detected. The maximum uptake rates, when recalculated

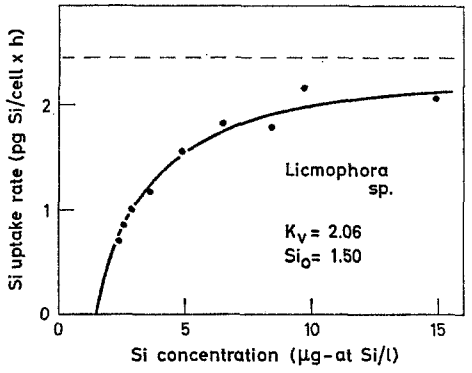

Fig. 7. *Licmophora* sp. Silicate-uptake rate as function of silicate concentration. Broken line: maximum rate

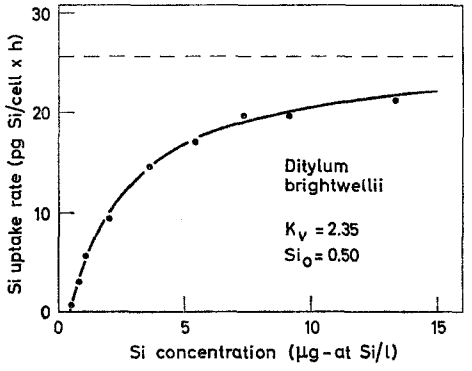

Fig. 8. *Ditylum brightwellii*. Silicate-uptake rate as function of silicate concentration. Broken line: maximum rate

highest one (*Thalassiosira decipiens*). The amount of residual silicate, Si_0, seemed to be greatest in species with a large K_v, with the exception of *Ditylum brightwellii*, in which Si_0 was small and K_v was large.

Also included in Table 1 are data on cell size and on cell silica content as measured in these diatoms when grown in a medium with ample silicate (Paasche, 1973, and in press). Again there was a clear trend, in that K_v tended to increase with increasing cell size (measured

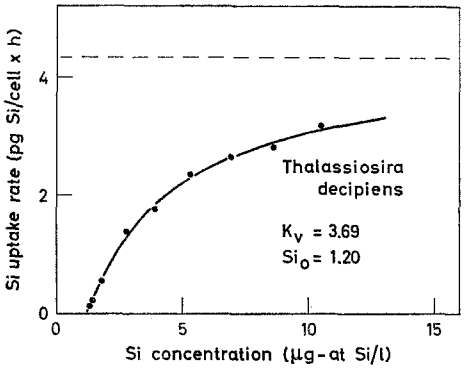

Fig. 9. *Thalassiosira decipiens*. Silicate-uptake rate as function of silicate concentration. Broken line: maximum rate

Table 2. *Mean values of the maximum silicate-uptake rate (V_{max}), calculated as doublings/day, and experimentally measured maximum growth rates (k_{max}) expressed in the same unit (data from Paasche, 1973, and in press). All data refer to 20°C and a saturating light intensity*

Species	V_{max} (doublings/day)	k_{max} (doublings/day)
Skeletonema costatum	0.48—0.85	2.4
Thalassiosira pseudonana	1.39	4.0[a]
Licmophora sp.	0.32—0.92	1.4
Ditylum brightwellii	1.01—4.32	3.2
Thalassiosira decipiens	0.43—0.93	1.3

[a] Computed from maximum specific growth rate (μ_{max}) in the chemostat according to the equation $k_{max} = \mu_{max}/0.69$.

as number of doublings per unit time (see below), indicated that only a small fraction of the cells divided and formed new valves within the 1 to 2 h allotted to incubation.

There was no way of deciding whether the first or the second uptake period in an experiment provided the better estimate of the kinetic constants, so no distinction was made between them in calculating the mean values of K_v, V_{max}, and Si_0 for each species. In Table 1, the 5 diatoms are arranged in the order of increasing K_v values. There was a four-fold difference between the lowest K_v (*Skeletonema costatum*) and the

either as cell diameter or as silica content). This may be the rule in nutrient uptake in general; thus, Eppley *et al.* (1969) observed a similar trend in nitrate and ammonia uptake experiments with a large number of marine plankton algae. There were some exceptions, however. The smallest K_v was observed in *Skeletonema costatum*, although the cells are somewhat larger than those of *Thalassiosira pseudonana*. Similarly, *T. decipiens* had the largest mean K_v of all, although the cells are smaller than those of *Ditylum brightwellii*. The statistical uncertainty of the data should be kept in mind. *Licmophora* sp., which is not strictly a plankton

alga and normally grows in the littoral zone, appeared to assimilate silicate with an efficiency comparable to that of the genuine plankton species.

The mean V_{max} values of Table 1 were recalculated according to an equation given by Eppley and Thomas (1969) to yield V_{max} expressed in the unit doublings/day:

$$V_{max}\,(\text{doublings/day}) = \frac{\log_2 10}{t} \cdot \log_{10}\left(\frac{P+p}{P}\right) \quad (2),$$

in which $t\,(= {}^1/_{24})$ is the incubation time in days; P the cell silicon content (data given in Table 1); and p the maximum amount of silicon taken up per cell during incubation (equal in magnitude to the V_{max} values in Table 1).

This allows a direct comparison between the silicate-uptake rates measured in the experiments and observed growth rates in the same species when grown with ample silicate (data from Paasche, 1973, and in press; see Table 2). It is evident from Table 2 that the calculated V_{max} rates are, on the whole, lower than would be expected from non-synchronous cell populations growing at an unrestricted rate. When nutrients such as phosphate or nitrate are re-added to nutrient-starved algal cultures, the initial (non-steady-state) uptake rates usually far exceed those during steady-state growth (Kuenzler and Ketchum, 1962; Eppley and Thomas, 1969). This, apparently, is not so with silicate.

Discussion

A meaningful discussion of nutrient-limited phytoplankton growth in nature presupposes that the steady-state growth kinetics are known. Many workers find it expedient to assume that growth rate varies with nutrient concentration according to a Michaelis-Menten curve. This facilitates discussion, since it is then possible to describe the efficiency with which low substrate concentrations can be used in terms of a half-saturation constant of growth.

Uptake experiments of the present kind are quick and simple to perform, and enable the investigator to accumulate much more data than do growth experiments. However, this advantage may be of dubious value, since there is a distinct possibility that the kinetics of nutrient uptake are different from those of nutrient-limited growth. Several possible reasons why this may be so have been discussed by Eppley and Thomas (1969). Among these, the most important, in the case of silicate at least, is that the amount of nutrient accumulated by a cell is not constant, but increases with increasing growth rate. In chemostat experiments with *Thalassiosira pseudonana* (Paasche, 1973), calculated half-saturation constants of steady-state silicate uptake were 3 to 4 times greater than those of growth; this was because the cell walls contained more silica in fast-growing than in slow-growing populations. The kinetics of nutrient uptake measured with nutrient-starved cells might, in turn, be different from those obtaining in the steady state. This is likely to happen particularly if intracellular nutrient stores that have become depleted are rapidly replenished during the initial uptake phase. There was little evidence that such was the case in the present experiments. The concept of a storage mechanism for silicon in diatoms was proposed by Werner and Pirson (1967), but may not be well founded in experimental facts (Darley and Volcani, 1969). The present mean value of 1.39 µg-at Si/l for the half-saturation constant of silicate uptake in *T. pseudonana* is almost exactly the same as the chemostat steady-state value (Paasche, 1973); but in view of the great spread and wide confidence limits of the individual K_v values (Table 1), this agreement may well be fortuitous.

Eppley and his coworkers (Eppley *et al.*, 1969; Eppley and Thomas, 1969) concluded that uptake experiments can be used, after all, to obtain the kinetic constants of nitrate-limited phytoplankton growth. The evidence for this was admitted to be open to criticism; and in any event it does not apply to the present experiments. It is interesting, however, that *Skeletonema costatum* (Fig. 1) and *T. pseudonana* (Fig. 2), both with a low K_v, showed a greater tendency towards the formation of thin shells during silicate shortage than did *T. decipiens* (Fig. 3) or *Licmophora* sp. (not shown; no similar infomation on variations in the cell silica-content could be obtained from *Ditylum* since the cells tended to throw off the cell walls after the cessation of net silicate uptake). This suggests that, even if the absolute values of the half-saturation constants of growth are much smaller than the K_v values of Table 1, the species can still be ranked in the same order of decreasing ability to use low silicate concentrations; in fact, the relative differences between the species might be larger than indicated by Table 1.

The remaining discussion will start from the assumption that the K_v values provide a correct measure of the relative (thought not of the absolute) magnitude of the half-saturation constants of growth. The residual silicate concentration, Si_0, will be ignored, since it cannot be excluded that it is an experimental artefact (Paasche, 1973).

Dugdale (1967) proposed that species with inherently low maximum growth rates may also exhibit small half-saturation constants of nutrient-limited growth. This would render them capable of outgrowing species with higher maximum growth rates but possessing less efficient nutrient-uptake mechanisms, especially towards the end of a bloom or in waters that are generally poor in nutrients. Kilham (1971), drawing upon the results of many investigators of fresh-water phytoplankton, considered that this model, when applied to silicate, could explain observed diatom succession-patterns. To see whether the present results might suggest a similar role for silicate in the marine

environment, growth rates of the 4 plankton species were calculated from an expression similar to Eq. (1), using the mean K_v values of Table 1, the maximum growth rates reproduced in Table 2, and relative values of $(Si - Si_0)$. The resulting curves are shown in Fig. 10.

The following conclusions are suggested by this diagram: (1) *Thalassiosira pseudonana* outgrows the other species, except at very low silicate concentrations where *Skeletonema costatum* is able to compete. (2) *T. decipiens* is outgrown by all others at all silicate concentrations; the low intrinsic growth rate of this

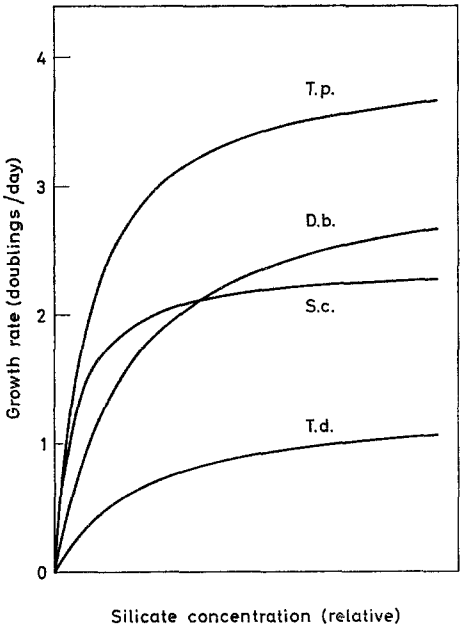

Fig. 10. Calculated growth rates as a function of relative silicate concentration for *Thalassiosira pseudonana* (T. p.); *Ditylum brightwellii* (D. b.); *Skeletonema costatum* (S. c.); *Thalassiosira decipiens* (T. d.). The growth rates refer to 20 °C and a saturating light intensity

species is not compensated for by an efficient silicate uptake mechanism. (3) *S. costatum* outgrows *Ditylum brightwellii* at low silicate concentrations, while the reverse is true at higher concentrations. Thus, in a natural succession induced by a gradual exhaustion of silicate, a *D. brightwellii* plankton would be followed by a *S. costatum* plankton. This is a rather hypothetical example, however, as *D. brightwellii* rarely forms a significant fraction of the phytoplankton biomass. In fact, this comparison of species may warrant no more than the very general statement that diatoms capable of forming large populations towards the end of a spring succession, or in the summer in temperate coastal waters, also seem to possess the ability to grow well at very low silicate concentrations. In Norwegian inshore waters, such as the Oslo Fjord (Braarud, 1945; Hasle and Smayda, 1960) and the Trondheim Fjord (Sakshaug, 1972), *S. costatum* frequently constitutes a major fraction of the phytoplankton in late spring or in summer, and the same may be true of *T. pseudonana* (Paasche, unpublished observation from the Oslo Fjord; this species is difficult to recognize in survey work, and there are few quantitative data on its distribution). The few silicate analyses available from these waters indicate that low concentrations may be found in spring or summer. There is indirect evidence that the diatom populations (consisting of *S. costatum* and other species) found in the Oslo Fjord in the summer season at times grow in water with a very low silicate content (Braarud, 1945; Hasle and Smayda, 1960).

Eppley *et al.* (1969), in their studies of nitrate and ammonia uptake by phytoplankton algae, were able to carry their analysis one step further, by including the known light requirements of some of the species. Ideally, an analysis of this sort should embrace both light and temperature effects, and probably a whole range of other factors as well, and the laboratory data should be compared with *in situ* growth rates of the plankton algae in a variety of natural situations. The present work does not even satisfy the relatively modest requirement that the selection of species should reflect the composition of natural plankton communities. However, the results do suggest that the role of silicon in marine ecology deserves greater attention than it has received so far. The rate of silicate uptake by plankton diatoms may become limited long before the supply of dissolved silicate in the sea water is exhausted, and low silicate concentrations may exert a selective influence on the species composition of diatom populations. In coastal waters the growth-regulating function of silicate may become accentuated wherever the concentrations of nitrogenous compounds and phosphates are raised through pollution. The possibility that the silicate supply governs the growth of diatoms in the polluted parts of the Oslo Fjord at certain times of the year will be tested by means of enrichment experiments and other studies that are at present being planned.

Summary

1. The following marine diatoms were grown in a medium with a low initial silicate concentration: *Skeletonema costatum*, *Thalassiosira pseudonana*, *T. decipiens*, *Ditylum brightwellii*, and *Licmophora* sp. Net silicate uptake stopped when there was 0.5 to 1.5 µg-at Si/l left in the medium; however, especially in cultures of *S. costatum* and *T. pseudonana*, growth continued, because the cells were able to reutilize silicate released by dissolution of older silica shells.

2. Short-term silicate uptake experiments were performed with the same 5 species. In all cases, the relationship between uptake rate and silicate concentra-

tion could be described by a Michaelis-Menten type of hyperbola, provided that a correction was made for the presence in the medium of reactive silicate that apparently could not be used by the diatoms. The magnitude of this correction was in the range of 0.3 to 1.3 µg-at Si/l (mean values for the different species).

3. The mean values of the half-saturation constants were as follows: in *S. costatum*, 0.80 µg-at Si/l; in *T. pseudonana*, 1.39 µg-at Si/l; in *Licmophora*, 2.58 µg-at Si/l; in *D. brightwellii*, 2.96 µg-at Si/l; in *T. decipiens*, 3.37 µg-at Si/l.

4. The maximum uptake rates were somewhat smaller than those expected to obtain in non-synchronous cultures growing at maximum growth rates.

5. The results suggest that low silicate concentrations may exert a selective influence on the species composition of phytoplankton populations in the sea.

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