chemical weathering processes. The suggestion is strongly supported by this study. The extent of chemical weathering and subsequent transport history of the sediment derived from even the same source materials may show appreciable variations in REE concentrations and in REE fractionation patterns. Cullers *et al.*[21] postulate that REE contents of clay minerals are determined primarily by the REE content of the source rock. However, at least in the granodiorite alteration profile, the weathering products contain REE concentrations appreciably different from the source rock. Cullers *et al.*[21] also noted an europium anomoly for the clays when normalised to NAS: no anomoly results from the weathering of the granodiorite. If the findings of this study are representative of subaerial weathering then the observed europium anomoly results from an europium anomoly of the source material or possibly from diagenesis through reduction of Eu^{3+} to Eu^{2+} (by carbonaceous material) and subsequent leaching of europium from the clays. In fact the wide variation of REE distributions found in the clay mineral groups studied by Cullers *et al.*[21] may, in large part, result from post-depositional processes.

Conclusions

I have shown that the REE can be mobilised in the supracrustal environment; that the enrichment and depletion of the REE are probably related to the *p*H of ground waters; that the fractionation of REE is probably a result of primary and secondary mineral abundances. The REE patterns of sediments and sedimentary rocks generally do not show the strong REE fractionation patterns observed in the altered materials studied here. The mechanical mixing of sedimentary detritus probably homogenises weathered materials to the extent that extreme fractionation patterns are not normally observed. Comparisons between this study and other studies suggest that subaerial and submarine weathering processes, and perhaps even hydrothermal processes, affect lanthanides similarly.

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1. Nance, W. B. & Taylor, S. R. *Geochim. cosmochim. Acta* **41**, 225–231 (1977).
2. Wildeman, T. R. & Condie, K. C. *Geochim. cosmochim. Acta* **37**, 439–453 (1973).
3. Taylor, S. R., & Hallberg, J. A. *Geochim. cosmochim. Acta* **41**, 1125–1129 (1977).
4. Balashov, Yu. A., Ronov, A. B., Migdisov, A. A. & Turanskaya, M. V. *Geochemistry Int.* **5**, 951–969 (1964).
5. Ronov, A. B., Balashov, Yu. A., & Migdisov, A. A. *Geochemistry Int.* **4**, 1–17 (1967).
6. Balashov, Yu. A. & Khitrov, L. M. *Geochemistry Int.* **4**, 404–407 (1967).
7. Burkov, V. V. & Podporina, Ye. K. *Dokl. Acad. Sci. USSR Earth Sci.* **177**, 214–219 (1966).
8. Wildeman, T. R. & Haskin, L. A. *Geochim. cosmochim. Acta* **37**, 419–438 (1973).
9. Haskin, L. A. & Schmitt, R A. in *Researches in Geochemistry* (ed. Abelson, P. H.) **2**, 234–258 (Wiley, New York, 1967).
10. Jacobs, J. W., Korotev, R. L., Blanchard, V. P. & Haskin, L. A. *J. Radio. Analyt. Chem.* **40**, 93–108 (1977).
11. Marsden, M. A. H. in *Geology of Victoria* (eds Douglas, J. G. & Ferguson, J. A.) 77–124 (Geol. Soc. Australia, Melbourne, 1976).
12. Pettijohn, F. J. *Sedimentary Rocks* 206–207 (Harper & Row, New York, 1975).
13. Garrels, R. M. & Christ, C. L. *Solutions, Minerals and Equilibria* 403–429 (Harper & Row, New York, 1965).
14. Garrles, R. M. in *Researches in Geochemistry* (ed. Abelson, P. H.) 405–420 (Wiley, New York, 1967).
15. Stumm, W. & Morgan, J. J. *Aquatic Chemistry* 171, 450, 485 (Wiley, New York, 1970).
16. Frey, F. A., Bryan, W. B. & Thompson, G., *J. geophys. Res.* **79**, 5507–5527 (1974).
17. Ludden, J. N. & Thompson, G. *Nature* **274**, 147–149 (1977).
18. Wood, D. A., Gibson, I. L., & Thompson, R. N. *Contr. Miner. Petrol.* **55**, 241–254 (1976).
19. Hellman, P. L. & Henderson, P. *Nature* **267**, 38–40 (1977).
20. Humphris, S. E., Morrison, M. A. & Thompson, R. N. *Chem. Geol.* **23**, 125–137 (1978).
21. Cullers, R. L., Chaudhuri, S., Arnold, B., Moon, L. & Wolf, C. W. *Geochim. cosmochim. Acta* **39**, 1691–1703 (1975).

Growth rate influence on the chemical composition of phytoplankton in oceanic waters

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The chemical composition of oceanic phytoplankton (by atoms) typically occurs in the proportions $C_{106}N_{16}P_1$. Yet, in laboratory growth conditions these proportions are only observed for marine phytoplankton at high growth rates when non-nutrient limitation is approached. Thus growth rates of natural phytoplankton populations in oceanic waters may be near maximal and hence non-nutrient limited. The uniformly low biomass and residual nutrient levels in such waters does not preclude the possibility of high growth rates because zooplankton grazing and nutrient regeneration within the euphotic zone may keep this highly dynamic system in a balanced state.

THERE is a large temporal and spatial variability of nutrient cycling in the surface waters of the oceanic environment[1]. Concomitant with the highly dynamic turnover of nutrients in oceanic surface waters are four characteristics of marine nutrient chemistry[2–5]. These are: (1) both phytoplankton biomass and aqueous nutrient concentrations are uniformly low in areas where there is little seasonal variation in the depth of the mixed layer; (2) the composition of particulate matter in marine waters is usually in the approximate proportions 106 : 16 : 1 (by atoms) for the three major elemental constituents, carbon, nitrogen and phosphorus (commonly referred to as the 'Redfield ratio'); (3) the dissolved inorganic N : P ratio of oceanic waters below the thermocline and in many coastal waters preceding bloom conditions is often about 16 : 1; (4) during bloom conditions inorganic nitrogen and phosphorus may disappear from solution in approximately a 16 : 1 ratio.

There are, however, numerous exceptions to these generalisations[3,4]. For example, within the euphotic zone phosphorus can be recycled more rapidly than nitrogen[6,7], so that a small residual of inorganic phosphorus is sometimes observed when inorganic nitrogen is undetectable[8]. Similarly, differential rates of nitrogen and phosphorus cycling preclude any *a priori* inferences as to the chemical composition of phytoplankton from the rates of disappearance of nutrients from surface waters[9]. Finally, particulate matter in surface waters contains varying amounts of non-phytoplankton material, ranging from a large percentage in nutrient-poor oceanic waters to a considerably smaller amount in productive coastal and upwelling systems[10].

There is, nevertheless, an abundance of data from recent literature on the chemical composition of phytoplankton in both productive and unproductive waters[6,11–17] to support the earlier conclusions of Redfield[2] and Fleming[18] that there is a striking consistency in the chemical composition of marine phytoplankton. For this discussion, however, considerable flexibility is allowed in the proportions of the chemical constituents without

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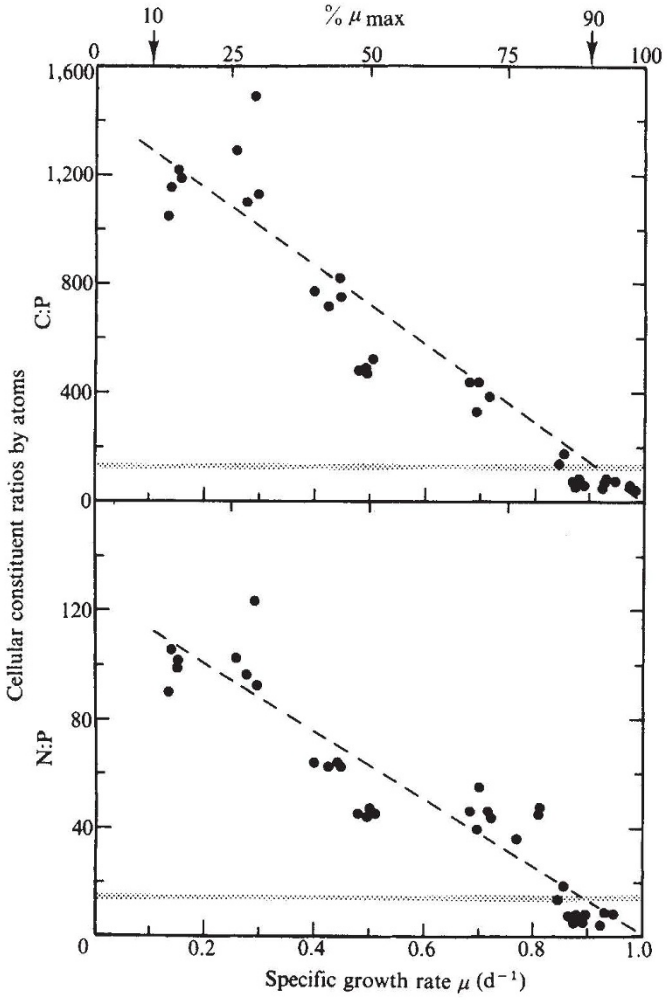

Fig. 1 The effect of specific growth rate on the carbon:phosphorus and nitrogen:phosphorus ratios of *Monochrysis lutheri* under phosphorus limitation in continuous culture at 18 °C and 0.03 cal cm^{-2} min^{-1} light intensity. Medium N:P ratios = 87:1 to 412:1. Shaded lines in Figs 1–3 represent C:P and N:P components of Redfield ratio ($C_{106}N_{16}P$). All dashed lines in these figures were drawn by eye and represent trends not absolute relationships between the respective cellular chemical ratios and growth rates. $\mu_{max} = 0.95\ d^{-1}$.

detracting from the generalisation of a uniform phytoplankton chemical composition. For example, values of ~75:1 to 150:1 for the C:P ratio and ~10:1 to 20:1 for the N:P ratio are consistent with the concept of a uniform chemical composition.

A clear distinction must be made between the processes controlling nutrient and phytoplankton interactions in productive coastal and upwelling waters and those in oceanic environments. We consider here only the latter system. By comparing the results of laboratory experiment on cultured marine phytoplankton with field observations, we propose a relationship between the nutrient-influenced growth rate and the elemental composition of oceanic phytoplankton.

Nutrient fluxes in the oceanic environment

On a long-term basis the dampening effect of the large ocean reservoir leads to a uniformity in the cycling of nitrogen and phosphorus. To maintain this consistency the major sources of nitrogen and phosphorus for phytoplankton growth in the euphotic zone of oceanic environments must be in balance with the major sinks of these nutrients on a long-term basis. Thus, fluxes of nutrients from vertical upward transport of nutrient-rich water across the thermocline, bacterial degradation of animal faeces and other detritus within the euphotic zone, and excretion of soluble nutrients by zooplankton and fish must be balanced by the assimilation of these nutrients by phytoplankton plus the loss of organisms and detritus to deep waters. Other nitrogen flux terms such as rain input, N_2 fixation and river discharges, and loss through permanent sediment burial and denitrification with evolution of N_2O and/or N_2, are essential components of a global nitrogen budget; but, at steady state these fluxes must cancel. In this regard, both rain and N_2 fixation provide an extremely small fraction of the nitrogen requirement for phytoplankton in the north central Atlantic[19], whereas river and waste water discharges are important sources of nitrogen and phosphorus in coastal waters[8]. But, because of the extent of the oceans, the overall impact of these latter additions on the global marine nutrient budget, and, in particular, on nutrient inputs to the oceanic mixed layer, has been negligible.

Mechanisms controlling marine nutrient chemistry

The generality of the Redfield ratio for phytoplankton and the 16:1 ratio for dissolved inorganic nitrogen and phosphorus in deep water leads to the apparent conclusion that nitrogen and phosphorus simultaneously influence phytoplankton growth in the oceans. But does the chemistry of the oceanic environment control the physiology and resulting chemical composition of the phytoplankton, or is the nutrient chemistry of the oceans manipulated by rather rigid physiological requirements?[4] The answer to this is not simple. A major complication is the often demonstrated fact, beginning with Ketchum's classical experiments on P-limited growth of *Nitzschia closterium*[20], that in laboratory conditions the chemical composition of phytoplankton can vary appreciably as a function of nutritional state and other growth conditions[16,21,22].

Growth rate effects on phytoplankton chemical composition

To establish the environmental conditions in which the chemical composition of marine phytoplankton can be characterised by the Redfield ratio, we grew three marine phytoplankton species in continuous culture: the chrysophyte *Monochrysis lutheri* under P-limitation and with NO_3^-–N as the nitrogen source, the diatom *Thalassiosira pseudonana* 3H under NH_4^+–N limitation, and the chlorophyte *Dunaliella tertiolecta* under NH_4^+–N, NO_2^-–N, NO_3^-–N, urea–N, and P-limitation. Details of the experimental procedures are reported elsewhere[23–25]. Although the

Table 1 Cellular constituent ratios of marine phytoplankton grown on varying nitrogen:phosphorus ratios and at different growth rates

Nutrient limitation	Species (clone)	Culture N:P ratio (atoms)	% μ_{max}	Cellular ratios (atoms) C:N:P	C:N
Phosphorus	*Monochrysis lutheri* (Mono)	87–412	10	1,300:115:1	11.3
			50	720:65:1	11.1
			90	106:15:1	7.1
	Dunaliella tertiolecta (Dun)	50	10	600:48:1	12.5
			50	325:32:1	10.2
Nitrogen	*Thalassiosira pseudonana* (3H)	5	10	63: 5:1	12.6
			50	68: 7:1	9.7
			90	106:15:1	7.1
	Dunaliella tertiolecta (Dun)	5	10	85: 5:1	17.0
			50	60: 5:1	12.0
			90	35: 5:1	7.0
		10	10	160:10:1*	16.0
			50	120:10:1	12.0
			90	70:10:1*	7.0
		15	10	300:15:1	20.0
			50	175:15:1	11.7
			90	106:15:1*	7.1

* Extrapolated from curves in Fig. 2.

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concentrations of nutrients in the media delivered to the growth chambers were high, the residual limiting nutrients in the cultures over most of the growth rate range remained at levels similar to those typical of oceanic mixed layers.

The effect of growth rate on the cellular C:P and N:P ratios was a distinct function of the degree and type of nutrient limitation. For P-limited growth of both *M. lutheri* and *D. tertiolecta* there was a large decrease in the cellular nutrient ratios from 600:1 to 1,000:1 for C:P and 50:1 to 100:1 for N:P at one end of the growth rate spectrum (~10% μ_{max}) to respectively 106:1 and 15:1 at the other end (~90% μ_{max}) (Figs 1, 2). In contrast, the cellular ratios varied quite differently for the N-limited cultures. For *T. pseudonana*, in which a single medium N:P ratio of 5:1 was used, the cellular ratios increased from 65:1 for C:P and 5:1 for N:P below 10% μ_{max} to respectively 120:1 and 15:1 above 90% μ_{max} (Fig. 3). In the case of N-limited growth of *D. tertiolecta*, there was no effect of either nitrogen source or growth rate on cellular N:P ratios, indicating complete assimilation of both nutrients (Fig. 2). On the other hand, the cellular C:P ratios all decreased with increasing growth rates. At 10% μ_{max} the C:P ratios were 300:1, 160:1, and 85:1 for the respective medium N:P ratios of 15:1, 10:1, and 5:1. The Redfield C:P ratio of 106:1 was approached at 90% and 60% of μ_{max} when the medium N:P ratios were respectively 15:1 and 10:1. In contrast, for a medium N:P ratio of 5:1 the cellular C:P ratio was always below the Redfield proportion and decreased to 35:1 at 90% μ_{max}.

Factors influencing the Redfield ratio

A common feature to the three growth experiments was that the Redfield ratio was approached only at high growth rates (Table 1). With the exception of the one *D. tertiolecta* experiment, this trend was true regardless of the medium N:P ratio. Based on the results of our previous studies[23–25], at 90% μ_{max} discernible nutrient limitation did not exist in any of the cultures. Hence, attainment of the Redfield ratio seem to coincide with conditions of near to complete nutrient saturation. In addition, the C:P and N:P ratios were considerably more affected by growth rate under P-limitation than when N was limiting, for example, 5–10-fold changes for P-limitation and only 2-3-fold variations for N-limitation (Table 1). As exemplified by the *M. lutheri* results, variations in the medium N:P ratio (87:1 to 412:1) did not affect the magnitude of the cellular N:P ratios when phosphorus was limiting. Yet, when nitrogen was limiting the magnitude of the medium N:P strongly influenced the resulting

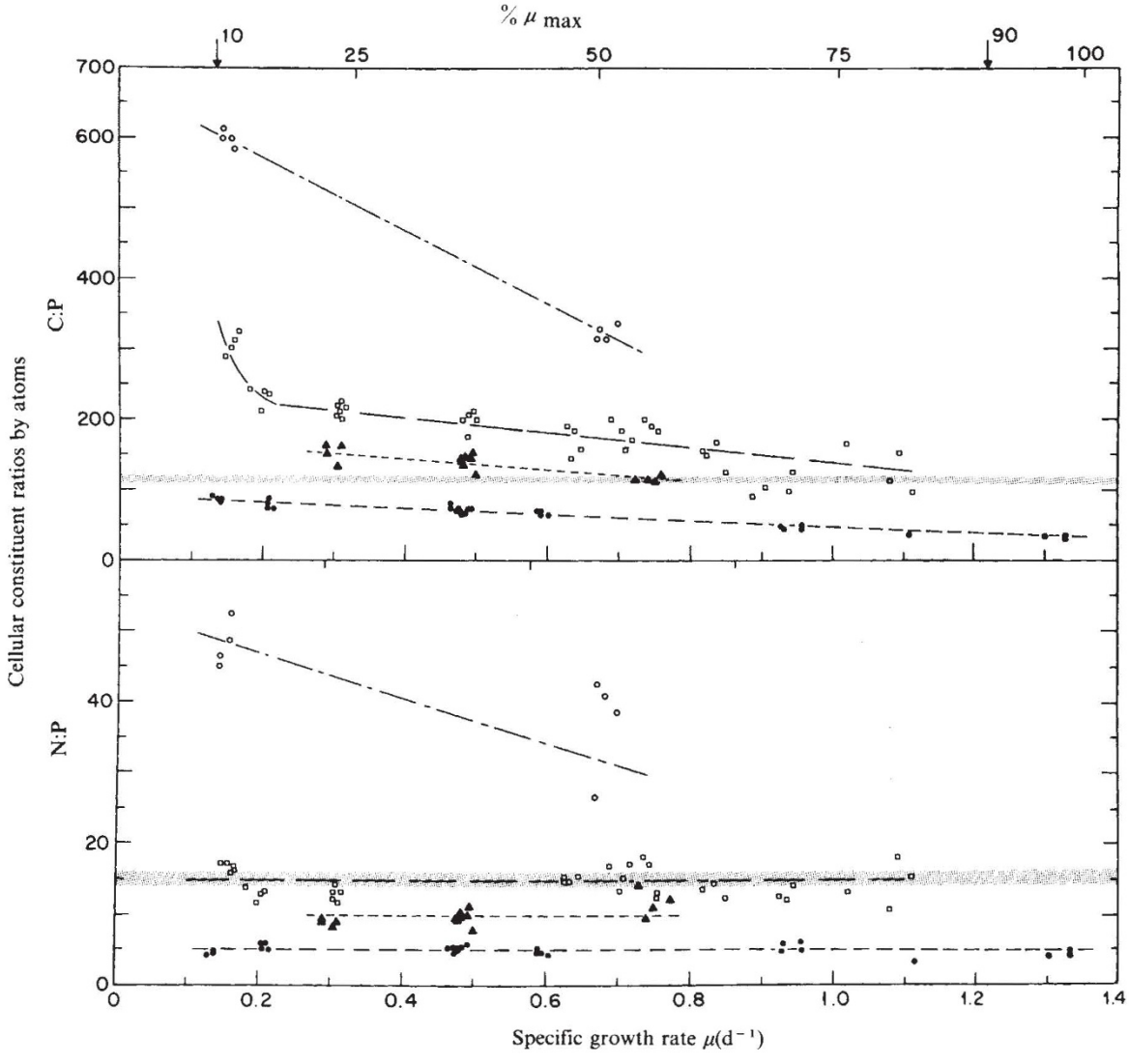

Fig. 2 The effect of specific growth rate on the C:P and N:P ratios of *Dunaliella tertiolecta* under phosphorus and nitrogen limitation in continuous culture at 19 °C and 0.06 cal cm^{-2} min^{-1} light intensity. Phosphorus limitation is presumed for medium N:P ≥ 50:1 and nitrogen limitation for N:P ≤ 15:1. ○, medium N:P = 50; □, medium N:P = 15; ▲, medium N:P = 10; ●, medium N:P = 5:1. The source of nitrogen had no affect on the relationships between cellular ratios and growth rates, as each cluster of four data points at a particular growth rate represents the four nitrogen sources used: NH_4^+-N, NO_2^--N, NO_3^--N, urea. $\mu_{max} = 1.35\ d^{-1}$.

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cellular N:P ratios. For *T. pseudonana* at low growth rates and for *D. tertiolecta* at any growth rate the medium and cellular N:P ratios were identical. In contrast, the C:P ratio typically approached the Redfield value only in the high growth rate regions. Similar results have been attained by other workers[16,21,26,27].

Biological effects

A possible explanation, therefore, as to why the chemical composition of natural marine phytoplankton is typically characterised by the Redfield ratio, is that growth rates in marine waters may be quite high. The corollary to this argument is that if phytoplankton growth rates are high, then nutrient availability is not the prime limiting factor in these waters.

Before examining the above hypothesis, it is instructive to consider first the available growth rate data for natural marine waters. Such measurements are difficult, and, as summarised in Table 2, the data are amazingly scant and conflicting; thus it is virtually impossible to draw any concrete conclusions as to the magnitude of *in situ* growth rates of oceanic phytoplankton. For example, Eppley[46] concluded that there was a direct correlation between the degree of eutrophy in marine waters and growth rates. Other workers[31,44,45], however, have found an opposite trend: higher growth rates associated with nutrient-poor and unproductive waters (Table 2). These contradictory conclusions are not amenable to close scrutiny because different procedures were used, and there is no completely satisfactory technique for making accurate estimates of *in situ* growth rates.

Hence, growth rates of marine phytoplankton are typically either <0.5 doublings per day ($\mu = 0.35\ d^{-1}$) or >1 doubling per day ($\mu = 0.69\ d^{-1}$)[32,34,44–46]. However, it is % μ_{max}, rather than the absolute value of μ that is germane to the question of the Redfield ratio. This point is exemplified by our observation that, although μ_{max} for the three species in the experiments described was distinctly different, the Redfield ratio was attained at growth rates close to μ_{max} in each case. The magnitude of μ_{max}, aside from being species specific, is also a function of temperature and light[46,47]. Therefore, growth rate data for natural populations, such as compiled in Table 2, provide no insight as to how fast these populations were growing relative to their potential maximum rates at the time of sampling.

The idea then that phytoplankton growth rates in oceanic waters are generally high and close to μ_{max} is at first difficult to accept, particularly when there is overwhelming evidence that nutrients in these waters are in such short supply and are often undetectable. To examine whether high growth rates and low nutrient levels can occur simultaneously, it is useful to consider the simple continuous culture as a dynamic system analogous to a segment of the open ocean. In such a system a limiting nutrient plus other nutrients in excess are added to the culture vessel at a fixed dilution rate (culture displacement per unit time). Cells assimilate the limiting nutrient and grow to a specific population size. Eventually a steady state is achieved in which the biomass produced in the culture per unit time is balanced by the discharge of biomass. An important result, often misunderstood, is that the growth rate of the organisms is then equal to the dilution rate, but it remains completely independent of the concentration of limiting nutrient in the medium feed. However, the steady state biomass is proportional to the concentration of added limiting nutrient[23].

In steady state conditions there seems to be a common trend of virtually undetectable residual limiting nutrient levels over the entire growth rate spectrum until just before the maximum growth rate is reached, regardless of which nutrient is limiting[23,24,48]. This common feature of marine phytoplankton physiology suggests that these organisms have a very high affinity for nutrients; thus at steady state it is possible to have simultaneously low or undetectable residual nutrient levels and high growth rates regardless of the biomass concentration.

On a long-term basis the euphotic zone of the open ocean probably approximates a continuous culture at steady state as well or better than any other aquatic system[32]: nutrient levels are uniformly low and the phytoplankton biomass is relatively invariant with time. Because nutrient input by vertical transport across the thermocline is generally considered to be small in oceanic systems (1–10% of the phytoplankton demand)[49,50], the main and preferred sources of nutrients (such as, NH_4^+, urea) are derived from animal excretion and bacterial mineralisation of detritus and dissolved organic matter[32,51,52]. At the same time phytoplankton biomass is continually cropped by zooplankton grazing. Hence, the zooplankton and bacteria act as the input and overflow mechanisms of the continuous culture[32]. The important question, however, for which there is no clear-cut answer is: at what rate does this dynamic system function?

The possibility that pelagic planktonic production rates could be near μ_{max} is compatible with observations of both low biomass and low residual nutrient levels, and with the notion that a particular nutrient limits the total biomass of the biological system rather than the actual rate processes. Cushing[53], in fact, argues that the euphotic zone of the open ocean is a relatively closed system, functioning at a very high rate with small temporal and spatial amplitudes in standing crop and nutrient levels; and it is the combination of zooplankton grazing and nutrient regeneration within the euphotic zone that is responsible for this tight coupling between trophic levels. Vertical transport of nutrients across the thermocline, although thought to be a minor source of nitrogen for phytoplankton[49,50], can at times be significant due to internal mixing[54]. Such pulsed inputs of nutrients to the mixed layer probably lead to proportional increases in standing crop, but, as shown above, do not necessarily affect the gross rate processes.

The microscale of nutrient cycling

Although the above arguments are appealing for describing a steady state oceanic system, we know from recent studies[55] that phytoplankton growth rates and transient nutrient uptake rates may be uncoupled. For example, we showed that *T. pseudonana* (3H) grown in continuous culture under modest NH_4^+-N limitation, can assimilate this nutrient at a rate up to 30 times its

Table 2 Summary of available phytoplankton growth rate data in doublings per day for natural marine waters

Location	Growth rate doublings per day*	Ref.
Sargasso Sea	0.26†	28
Florida Strait	0.45†	28
Carolina Coast	0.37†	28
Montauk Pt, L.I.	0.35†	28
S Calif. Coast	0.25–0.4†	29
S Calif. Coast	0.7†	30
NW Atlantic	0.2–1.7	31
N Pacific	0.2–0.4	32
Northern N Pacific	0.36–0.89	33
North Sea	0.67–1.33	34
Sargasso Sea	0.05–0.14	35
Tyrrhenian Sea	0.07–0.25	35
Baja, Calif. Coast	0.2–1.4	36
Peru Current	0.67†	37
Peru Current	0.73†	38
SW Africa Coast	1.0†	39
W Arabian Sea	>1.0†	40
Narragansett Bay, RI	0.4–1.94	41
Narragansett Bay, RI	<0.1–3.8	42
Santa Monica Bay, Calif.	0.3–0.7	43
Sargasso Sea	8	44
Oligotrophic Waters	6.6	45
Mesotrophic Waters	2.3	45
Eutrophic Waters	0.14	45

No distinction is made between coastal, upwelling and oceanic waters. The lack of correlation between growth rate and degree of productivity attests to the confusion regarding growth rate measurements and their meaning.

* Doublings per day × 0.693 = specific growth rate μ.

† Obtained from Table 2 in ref. 46.

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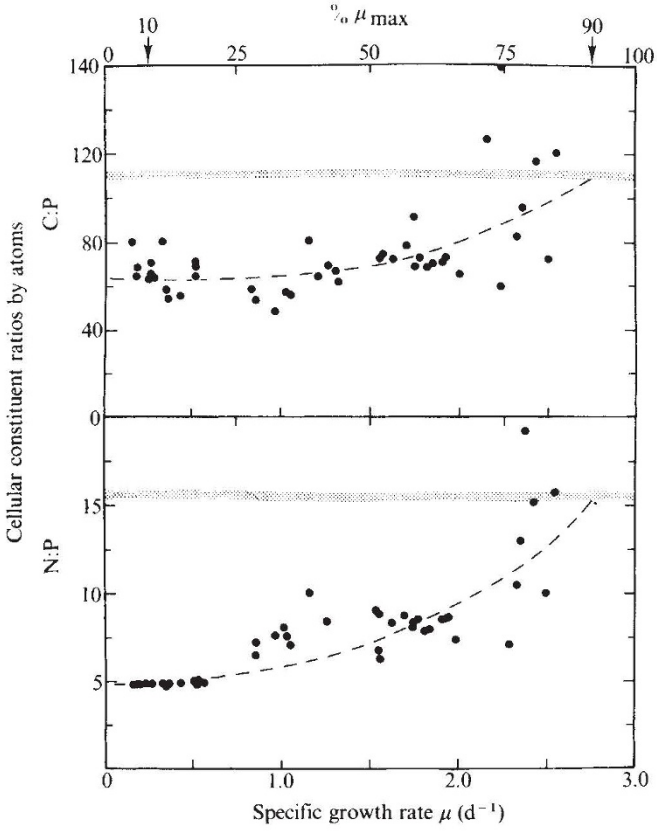

Fig. 3 The effect of specific growth rate on the C:P and N:P ratios of *Thalassiosira pseudonana* 3H under ammonium limitation in continuous culture at 19 °C and 0.06 cal cm^{-2} min light intensity. Medium N:P ratio = 5:1. $\mu_{max} = 3.02\ d^{-1}$.

specific growth rate when exposed to saturating concentrations of NH_4^+ for short (5 min) periods[55]. The major implication of this finding is that a nutrient-stressed phytoplankton cell need be exposed to an elevated nitrogen concentration for only a small fraction of its doubling to obtain a significant ration of this nutrient. Similar data for phosphorus uptake are available, indicating that the dynamics of phosphorus assimilation are similar to those for nitrogen[56].

This situation could exist on a microscale in which a phytoplankton cell randomly and perhaps frequently passes within a zone of elevated NH_4^+, urea, or PO_4^{-3} concentration surrounding a zooplankter or bacterial assemblage. This hypothesis is consistent with the observation that nutrient concentrations in oceanic surface waters are frequently below detectable levels when, on the basis of measured photosynthetic rates, there is no indication of serious nutrient limitation[55]. From this perspective, the response of an individual oceanic phytoplankter is quite unlike that of the total population maintained at steady state in a continuous culture. By enhanced nutrient uptake the nutrient-stressed cell can rapidly attain the nutrient ration necessary to synthesise cellular material at maximal rates. A more complete understanding of the nutrition of these organisms in nature will require laboratory culture procedures which mimic the short-term response of the individual cell to a transient nutrient regime.

Although these processes actually occur on a temporal and spatial scale that we cannot observe, the net sum of all these events appears as a 'steady state' ocean system, but on a much larger scale. Thus the hypothesis that the ocean system is in a highly dynamic state, characterised by high rates of phytoplankton growth and nutrient turnover, is entirely consistent with our experimental results showing attainment of the Redfield ratio when phytoplankton growth rates and growth rate potential are close to μ_{max} at essentially undetectable nutrient concentrations. Unfortunately, the major limitation of this hypothesis is that the supporting evidence is highly circumstantial. Only when accurate measurements of *in situ* growth rates are possible will the question be answered adequately.

Conclusions

The possibility that the N:P ratio of 16:1 in the bulk of the world's oceans is the result of a combination of geochemical processes controlling the input of phosphorus and microbial processes such as N_2 fixation, nitrification, and denitrification controlling nitrogen inputs cannot be discounted. That phytoplankton can simultaneously strip nitrogen and phosphorus from solution when the aqueous N:P ratio varies from 5:1 to 15:1, is consistent with the hypothesis that the chemical composition of marine phytoplankton is strongly influenced by the chemistry of the surrounding waters, irrespective of growth rate. Yet, the concept of geochemical control of the chemical composition of phytoplankton does not explain why inorganic carbon, which is present in quantities far in excess of the requirements of marine phytoplankton, appears to be assimilated in the Redfield proportions only at high growth rates (Table 1). This effect can be seen most clearly by observing the C:N ratios under N-limitation (Table 1). Whereas the N:P ratios are relatively invariant over the growth rate spectra (Figs 1–3), the C:N ratios, particularly those for *D. tertiolecta*, vary significantly. Clearly the carbon component of the Redfield ratio is controlled primarily by physiological and not aqueous chemical factors.

Our results demonstrate the circumstances in which the Redfield ratio can be attained, and point to the possibility that the rate processes in nutrient impoverished oceanic regions may be in a highly dynamic and balanced state. One conclusion that can be stated with some certainty is that severe phosphorus limitation probably does not exist in the world's oceans. Otherwise the C:P ratios of phytoplankton would be far larger than the typically observed values of 75:1 to 150:1. However, to explore further these questions involves new approaches for studying microbial interactions on temporal and spatial scales that are far smaller than were previously assumed to be important.

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1. Dugdale, R. C. in *Modelling of Marine Systems* (ed. Nihoul, J. C. J.) 187 (Elsevier, Amsterdam, 1975).
2. Redfield, A. C. in *James Johnstone Memorial Volume* (ed. Daniel, R. J.) 176 (Liverpool University Press, 1934).
3. Ketchum, B. H. *Ecol. Monogr.* **17**, 309 (1947).
4. Redfield, A. C. *Am. Scient.* **46**, 205 (1958).
5. Corner, E. D. S. & Davies, A. G. *Adv. mar. Biol.* **9**, 101 (1971).
6. Antia, N. J., McAllister, C. D., Parsons, T. R., Stephans, K. & Strickland, J. D. H. *Limnol. Oceanogr.* **8**, 166 (1963).
7. Corner, E. D. S., Head, R. N. & Kilvington, C. C. *J. mar. biol. Ass. UK* **52**, 847 (1972).
8. Ryther, J. H. & Dunstan, W. M. *Science* **171**, 1008 (1971).
9. Banse, K. *Deep Sea Res.* **52**, 762 (1974).
10. Menzel, D. W. in *The Sea—Marine Chemistry* Vol. 5 (ed. Goldberg, E. D.) 659 (Interscience, New York, 1974).
11. McAllister, C. D., Parsons, T. R. & Strickland, J. D. H. *J. Cons. Int. Explor. Mer.* **25**, 240 (1960).
12. McAllister, C. D., Parsons, T. R., Stephans, K. & Strickland, J. D. H. *Limnol. Oceanogr.* **6**, 237 (1961).
13. Parsons, T. R. & Strickland, J. D. H. *J. Fish. Res. Bd Can.* **18**, 1001 (1961).
14. Strickland, J. D. H., Holm-Hansen, O., Eppley, R. W. & Linn, R. J. *Limnol. Oceanogr.* **14**, 12 (1969).
15. Hang, A., Myklestad, S. & Sakshaug, E. *J. exp. mar. Biol. Ecol.* **11**, 15 (1973).
16. Perry, M. J. *Limnol. Oceanogr.* **21**, 88 (1976).
17. Sen Gupta, R., Sankaranarayanan, V. N., De Sousa, S. N. & Fondekar, S. P. *Ind. J. mar. Sci.* **5**, 58 (1976).
18. Fleming, R. H. *Proc. 6th Pac. Sci. Congr.* 1939, **3**, 535 (1940).
19. Carpenter, E. J. & McCarthy, J. J. *Limnol. Oceanogr.* **20**, 384 (1975).
20. Ketchum, B. H. *J. Cell. comp. Physiol.* **13**, 373 (1939).
21. Fuhs, G. W., Demmerle, S. D., Canelli, E. & Chen, M. *Am. Soc. Limnol. Oceanogr. Spec. Symp.* **1**, 113 (1972).
22. Sakshaug, E. & Holm-Hansen, O. *J. exp. mar. Biol. Ecol.* **29**, 1 (1977).
23. Goldman, J. D. *J. Phycol.* **13**, 251 (1977).

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24. Goldman, J. C. & McCarthy, J. J. *Limnol. Oceanogr.* **23,** 695 (1978).
25. Goldman, J. C. & Peavey, D. G. *Appl. envir. Microbiol.* (in the press).
26. Rhee, G. *J. Phycol.* **10,** 470 (1974).
27. Rhee, G. *Limnol. Oceanogr.* **23,** 10 (1978).
28. Riley, G. A., Stommel, H. & Bumpus, D. F. *Bull. Bingham Oceanogr. Collect. Yale Univ.* **12** (1949).
29. Eppley, R. W. *et al. Calif. Mar. Res. Commun., CalCOFI Rep.* **16,** 74 (1972).
30. Eppley, R. W., Reid, F. M. H. & Strickland, J. D. H. *Bull. Scripps Inst. Oceanogr. Univ. Calif.* **17,** 33 (1970).
31. Sutcliffe, W. H., Sheldon, R. W. & Prakash, A. *J. Fish. Res. Bd Can.* **27,** 1917 (1970).
32. Eppley, R. W., Renger, E. H., Venrick, E. L. & Mullin, M. M. *Limnol. Oceanogr.* **18,** 534 (1973).
33. Saino, T. & Hattori, A. *Mar. Sci. Commun.* **3,** 1 (1977).
34. Cushing, D. H. *Trans. R. Soc. S. Afr.* **40,** 17 (1971).
35. Swift, E. & Durbin, E. G. *Deep Sea Res.* **19,** 189 (1972).
36. Smayda, T. J. *Deep Sea Res.* **22,** 151 (1975).
37. Strickland, J. D. H., Holm-Hansen, O., Eppley, R. W. & Linn, R. J. *Limnol. Oceanogr.* **14,** 23 (1969).
38. Beers, J. R., Stevenson, M. R., Eppley, R. W. & Brooks, E. R. *Fish. Bull. U.S.* **69,** 859 (1971).
39. Hobson, L. A. *Invest. Pesq.* **35,** 195 (1971).
40. Ryther, J. H. & Menzel, D. W. *Deep Sea Res.* **12,** 199 (1965).
41. Durbin, E. G., Krawiec, R. W. & Smayda, T. J. *Mar. Biol.* **32,** 271 (1975).
42. Smayda, T. J. *Norw. J. Bot.* **20,** 219 (1973).
43. Weiler, C. S. & Chisholm, S. W. *J. exp. mar. Biol. Ecol.* **25,** 239 (1976).
44. Sheldon, R. W. & Sutcliffe, W. H. *Limnol. Oceanogr.* **23,** 1051 (1978).
45. Koblentz-Mishke, O. J. & Vedernikov, V. I. *Mar. Sci. Commun.* **2,** 357 (1976).
46. Eppley, R. W. *Fish. Bull.* **70,** 1063 (1972).
47. Goldman, J. C. & Carpenter, E. J. *Limnol. Oceanogr.* **19,** 756 (1974).
48. Eppley, R. W. & Renger, E. H. *J. Phycol.* **10,** 15 (1974).
49. Emery, K. O., Orr, W. L. & Rittenberg, S. C. in *Essays in Natural Science in Honor of Captain Allan Hancock,* 299 (University of Southern California Press, Los Angeles, 1955).
50. Broecker, W. S. *Chemical Oceanography* (Harcourt Brace Jovanovich, 1974).
51. Dugdale, R. C. & Goering, J. J. *Limnol. Oceanogr.* **12,** 196 (1967).
52. McCarthy, J. J. *Limnol. Oceanogr.* **17,** 738 (1972).
53. Cushing, D. H. in *Marine Ecology and Fisheries* (Cambridge University Press, 1975).
54. McGowan, J. A. & Hayward, T. L. *Deep Sea Res.* **25,** 771 (1978).
55. McCarthy, J. J. & Goldman, J. C. *Science* **203,** 670 (1979).
56. Burmaster, D. M. thesis, MIT, Cambridge, 1978).

A mutation altering the function of a carbohydrate binding protein blocks cell–cell cohesion in developing *Dictyostelium discoideum*

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In Dictyostelium discoideum, *carbohydrate binding proteins (CBPs) or lectins have been implicated in the molecular basis of cellular cohesion. To determine the role of these CBPs, we have attempted to isolate structural gene mutants in which the CBPs have a defective affinity for carbohydrate ligands. We now report the isolation of a spontaneous, cross-reacting material (CRM) mutant which is non-cohesive and fails to develop. The mutant seems to have a defect in the structural gene for one of the two developmentally regulated carbohydrate binding proteins (CBP-26), which renders it unable to bind to galactose-containing ligands. The fact that wild-type cells interact with the mutant and carry it through development strongly supports a model of cell–cell interaction in which cohesion is mediated by complementary molecules.*

AN important goal of biology is an understanding of the molecular basis of the cell–cell interactions which underlie morphogenesis and development. In several developmental systems, specific molecules have been proposed as possible mediators of cell–cell adhesion[1–8]. The candidate molecules are usually said to mediate specific cellular adhesions because either the purified molecule causes cell aggregation[3,9–17], or antibodies against the molecules block cellular adhesions or even development[18–24]. However, although intriguing, these studies are indirect. Similarly, in the cellular slime mould *Dictyostelium discoideum*, our own studies and those of Rosen *et al.*[25] have defined two developmentally regulated carbohydrate binding proteins (CBPs) which have been suggested to be involved in specific cell–cell adhesion. The CBPs are developmentally regulated and appear on the cell surface when the cells become cohesive[25,26]. In their native form they are homotetramers, with molecular weights of about 100,000. On reduction and separation of their subunits in SDS–acrylamide gels, the two CBPs can be distinguished from each other by the molecular size of their subunits [26,000 (CBP-26) and 24,000 (CBP-24) (ref. 27)]. Structural and kinetic studies have shown that CBP-26 and CBP-24 are distinct gene products with a very similar time course of appearance during development[27–29]. However, as for the candidate molecules of other systems, the evidence that the CBPs of *D. discoideum* have a role in development is largely indirect[30].

We now report the isolation of a spontaneous cross-reacting material (CRM) mutant which seems to have a defect in the structural gene for CBP-26. The mutant is non-cohesive, suggesting a critical role for CBP-26 in cellular cohesion and subsequent development. The finding that when mutant cells are mixed with wild-type cells, the mutant develops normally (synergy), is strong evidence for a model ('lectin-ligand') in which complementary molecules mediate specific cell–cell cohesion.

Isolation of HJR-1

A population of NC-4 cells was enriched for cells lacking CBPs by affinity chromatography on Sepharose 6MB coupled to desialated fetuin. Whereas wild-type cells bind to the galactose matrix through their CBPs, cells lacking CBPs do not bind (J.R., unpublished). Wild-type NC-4 cells were collected after 16 h of development (when the amount of cell surface CBPs is maximal), and 10^8 cells were chromatographed on a 4-ml Sepharose 6MB column. About 0.3% of the cells came out during column washing. Two hundred of the non-binding cells were plated out in association with *Klebsiella aerogenes* and allowed to grow and develop at 22 °C. One variant plaque that failed to develop normally was picked for further study and purified by successive single-plaque isolations; the mutant strain was designated HJR-1. (A detailed genetic study of this and other mutants obtained by this technique and a full description of the use of affinity chromatography to isolate CRM mutants of *D. discoideum* will be published elsewhere (J.R. *et al.*, in preparation).)

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