Life between skylla and charybdis; microbes in a sea of competition and predation

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ABSTRACT

Investigations in marine microbial ecology have tended to be restricted in scope to subdiciplines, such as phytoplankton ecology, bacteriology, or protozoology, a fact most probably a result more of historical traditions in educational systems, rather than from the potential for any of one of these subdiciplines alone to fully explain the mechanisms regulating the role of any of one microbial group in the ecosystem. We discuss how the control of bacterial production in the pelagic photic zone can be understood as a combination of competitive, commensal, and predatory mechanisms, involving phytoplankton, bacteria, and protozoa in a tightly coupled network. With particular focus on the Mediterranean, we discuss how such an approach can explain features observed in phosphate deficient systems.

The prolonged existence of a population in an ecosystem requires that, over time, growth balance losses. This may seem self-evident, yet investigations of the dynamics of populations often tend to focus on only one aspect of these processes, such as growth conditions determined by competition for food or losses due to predation or viral lysis. The importance of considering both processes simultaneously can be illustrated by a simple four population model (Fig. 1) with heterotrophic flagellates preying on bacteria, ciliates preying on auto- and heterotrophic flagellates, and bacteria competing with autotrophic flagellates for mineral nutrient [8]. In the Mediterranean, investigations both in the northwest [10, 11] and eastern basin [12] indicate that growth rates of both phytoplankton and heterotrophic bacteria can be simultaneously P-limited. From theoretical arguments, bacteria, with their high surface:volume ratio, are expected to be the superior competitors for free phosphate at permanently low phosphate concentrations. Considering competition only, this leads to the following variation of Hutchinson's paradox : *Why do bacteria not outcompete phytoplankton until primary production decreases to a level where bacteria become C-limited?*

While there are many theoretical solutions to Hutchinson's paradox, perhaps the simplest version is that illustrated in Fig.1. With a sufficiently efficient predator keeping the biomass of the superior competitor in check, the theoretical problem vanishes. The otherwise winning competitor is then prevented from incorporating all of the available P its biomass, and there is a resulting steady state solution for the system in which all competitors and predators can coexist simultaneously.

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Using radio-labelled orthophosphate, combined with size-fractionation on polycarbonate filters, one can estimate the fraction (f) of orthophosphate taken up by bacteria. In studies of surface water in Villefranche Bay on the French Mediterranean coast, f was typically found to be ≈ 50% [2]. An observed tendency for f to decrease through the autumn period of the study was presumably related to destabilisation of the water column caused by cooling of the top water layer [2]. In a Norwegian fjord, f was found to vary between 10 and 90%, with high values of f correlated with short turn-over times of orthophosphate in the brackish layer [9].

If we make the simplifying assumption that the concentration of bioavailable phosphate ($S_{n)}$ is low enough that uptake is proportional to S_n, the flow of phosphate into bacteria (V_B) is the product of three factors, i.e., $V_B = \alpha_B S_n B$, where α_B is the bacterial specific affinity for phosphate (slope of the uptake curve as substrate concentration approaches 0), and B is bacterial biomass. The distribution of phosphate uptake between phytoplankton and bacteria is thus a function, not only of their relative abilities to sequester phosphate (i.e., the ratio $\alpha_B:\alpha_A$, where α_A is the phytoplankton affinity), but also of the ratio of bacterial to phytoplankton biomass (B:A), and thus the ability of predators to keep the biomass of bacteria and phytoplankton in check.

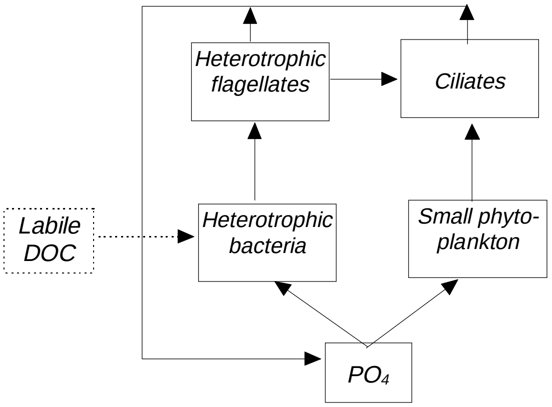

Fig. 1. Idealized food web illustrating P-flow (solid lines) and bacterial consumption of labile DOC (dotted line), allowing coexistence of P-limited bacteria and phytoplankton due to predation by protozoa. Distribution of phosphate uptake between P-limited bacteria and phytoplankton is, in this structure, determined both by their relative affinities for phosphate and by their relative biomasses.

Based on combining an estimate of the loss rate of bacteria with particulate-P concentration in the biomass size-fraction 0.2-1 μm, the concentration of bioavailable orthophosphate in both Villefranche Bay, NW Mediterranean [7] and in Sandsfjorden, Norway [9] was estimated to be ca. 1 nM. Assuming diffusion limitation, i.e., all substrate molecules contacting the cell are captured, the specific affinity constant of a perfectly spherical cell will be $3D/\sigma r^2$ [6], where D, σ, and r are the diffusion constant for orthophosphate, volume specific phosphorus content of the cell, and the cell radius, respectively. Using $D = 10^5\ cm^{-2}\ s^{-1}$ as a typical value for small molecules in water, a cell radius of 1 μm, a specific carbon content of $2.4\ 10^{-16}$ mol-P μm^{-3} (corresponding to a specific gravity of 1.2, 25% of wet weight as C, and a molar C:P-ratio of 106 in phytoplankton), gives $\alpha_A \approx 0.046$ l nmol-$P^{-1}\ h^{-1}$. At a substrate concentration of 1 nM P l^{-1}, this corresponds to a phytoplankton growth rate of $\mu_A \approx \alpha_A S_n = 0.046\ h^{-1} = 1.1\ d^{-1}$.

Interestingly, this corresponds to the growth rate estimated for the *Synechococcus* population in Villefranche Bay from flow cytometric analyses [5]. The data thus suggests that, in these P-deficient environments, cells in the size range of pico-autotrophs may be diffusion-limited.

Heterotrophic bacteria have an advantage in this game due to their smaller size, but they also may have a disadvantage due to their high specific P-content [4]. Assuming a spherical bacterial cell of radius 0.3 μm and a specific P-content of $5\ 10^{-7}$ nM-P μm^{-3} (same assumptions as for phytoplankton, but assuming a molar C:P-ratio of 50 [4]), gives a theoretical $\alpha_B \approx 0.24$ l nM-P^{-1} h^{-1}, i.e. a relative advantage over phytoplankton by a factor of ca. 5. We have tried to estimate α_A and α_B for natural, mixed, populations. Preliminary results (data not shown), based on chlorophyll and total fluorescence microscopy counts as estimates of phytoplankton and bacteria biomass respectively, give phytoplankton affinities very similar to those calculated theoretically above, while the bacterial advantage seems smaller with an estimated ratio α_B:α_A about 1 [Thingstad, these Proceedings]. Precise estimation is difficult, however, not only due to the problem of obtaining exact biomass estimates in phosphorus units, but also to the problem of determining the fraction of bacteria actively involved in phosphate uptake. If a high percentage of the bacteria counted are assumed to be dead or inactive [13], the estimates of bacterial affinity would be larger. When bacterial growth rate is P-limited in the system diagrammed in Fig. 1, bacterial carbon demand will be proportional to the second power of ciliate biomass (subject to assumptions on proportionality between food concentration and food uptake, and on equilibrium in parts of the system) [Thingstad, this Proceedings]. The results are then consistent if the sum of autochthonous production and allochthonous input of organic substrates exceeds this carbon demand. If production is smaller than this, the pool of degradable organic-C would sooner or later be depleted, in which case bacterial growth rate should shift from P to C limitation. With a food web structure as shown in Fig.1, the ammensal relationship created by nutrient limited phytoplankton excreting organic substrates for bacterial growth [1] would not lead to bacteria out competing phytoplankton because a high production of organic-C would force the system into P-limitation of the bacteria.

The idealised structure shown in Fig. 1 implies that all phytoplankton are subject to predation by ciliates. In reality, phytoplankton can avoid this by increasing their size. The price for this may seem excessive however, since maximum affinity decreases as the second power of cell size. Since loss due to sinking also increases as the second power of cell size (Stoke's law), compensating mechanisms may be necessary to allow such a strategy to be used for predator avoidance. An interesting suggestion is that diatoms, with their large vacuole, can actually compensate for having a large size by having a small volume-specific content of the limiting mineral nutrient [3]. As a consequence, the specific activity of diatoms would be less than anticipated based on their size alone. Apparently the price to be paid for this compensating strategy is the obligate diatom requirement for silicate necessary to build the rigid frustules.

If we expand the food web structure of Fig. 1 to the one shown in Fig.2 by adding diatoms and copepods, the suggestion from the above argument is that the size distribution of phosphate uptake may be more a function of predator control of the biomass of bacteria and of phytoplankton in different size classes, than a consequence of large differences in

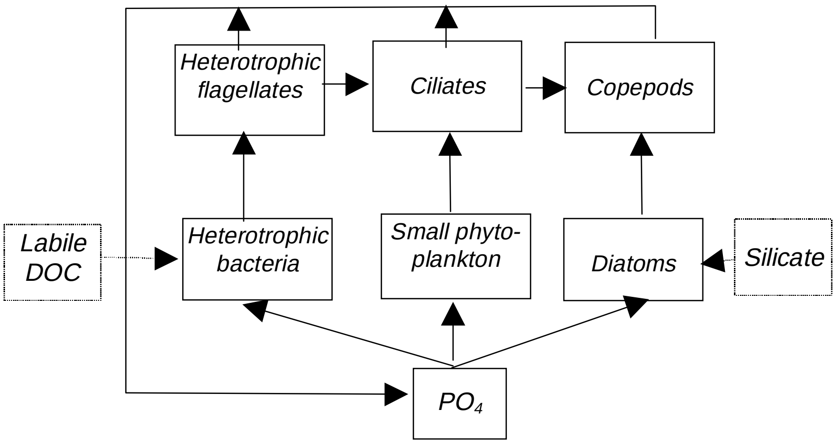

Fig. 2. Idealized food web structure expanded from Fig.1 by adding diatoms and copepods. Note the symmetrical role of labile DOC and silicate in their potential to restrict P-flow through the left "microbial" and the right "classical" side of the food web, respectively.

their specific affinities for phosphate. Another interesting feature of this food web structure is the symmetrical role suggested for labile DOC and silicate. A maximum spread in the size distribution of phosphate uptake exists in the case where labile DOC and silicate are in excess of bacterial and diatom demand, respectively. Depletion of labile DOC will act like a choke for the P-flow through the left 'microbial' side of system, while lack of silicate will have a similar effect on the P-flow through the right 'classical' side. Conversely, one would expect that stimulating diatom growth by adding silicate to a Si-depleted system should reduce bacterial degradation of labile DOC due to increased competition for phosphate. Experimental data from mesocosm experiments (data not shown) so far seem to confirm this expectation.

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