NPo-forskning fra Miljøstyrelsen

Nr. C5 1990

Carbon Dynamics in Coastal Marine Environments

Miljøministeriet **Miljøstyrelsen**

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Bo Riemann, Peter Koefoed Bjørnsen,
Torkel Gissel Nielsen, Steen Jesper Horsted

The Water Quality Institute
Marine Biological Laboratory, University of Copenhagen
Danish Institute for Fisheries and Marine Research

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Indholdsfortegnelse.

Dansk resume.

Det var projektets overordnede formål at integrere kemiske og biologiske parametre i en analyse af regulerende faktorer i stofomsætningen i kystnære marine områder. Det daværende videngrundlag var hovedsageligt bygget på traditionelle regressionanalyser af kvælstof og fosfor samt primærproduktion, og anvendelsen af disse informationer til at forudsige ændringer i de kystnære områder er meget dårlig. Vores resultater og konklusioner repræsenterer et første trin i en integreret analyse af de kystnære marine områders struktur og funktion. Vi har udarbejdet en række foreløbige kulstof budgetter og har kunnet vise, hvorledes tilsætning af næringsstoffer påvirker udvikling af biomasse og processer mellem autotrofe (planter) og heterotrofe (dyr og bakterier) organismer, og hvorledes biologiske processer som muslingers filtrering og zooplankton og fisks predation påvirker stofomsætningen. Eksempelvis viste det sig, at tilsætning af næringsstoffer øgede flowhastigheden af organisk stof igennem bakterier og heterotrofe flagellater og mindskede systemets stabilitet f.eks. ved pludselige ændringer i iltforbruget. Vi kunne sammenfattende vise, at balancen mellem tilsætning af næringsstoffer, planteplanktonets størrelsesfordeling, muslingernes fysiologiske tilstand og forekomst af fisk, der æder plankton, er afgørende for de kystnære marine områders struktur og funktion.

Resume.

The overall objective of this project was to examine important factors regulating carbon dynamics in coastal marine environments. The present understanding of the processes leading to deterioration of water quality is gene-

rally weak and often exclusively based on mass balances of the nitrogen and phosphorus cycles. Experience has shown that the predictive value of simple relationships between nitrogen and phosphorus is nevertheless limited. We tested the hypothesis that biological processes as well could have an impact on the production and decomposition of organic matter. While the project's results do not allow us to present a complete evaluation of factors regulating the carbon dynamics, we have demonstrated a provisional scenario of the carbon budget. We have also presented evidence concerning the effects of nutrient additions on the biomasses of and processes between autotrophs (plants) and heterotrophs (animals and bacteria) in the food web and the effects of biological mechanisms, such as filtration by mussels, grazing by zooplankton and predation by fish control on the structure and functions of the ecosystem. Moreover, we demonstrated how nutrient additions increased the flux of organic material through heterotrophic microorganisms (bacteria and heterotrophic flagellates) thereby decreasing the stability of the ecosystem. In summary, the project demonstrated that the balance between nutrient additions, the size distribution of phytoplankton, the physiological conditions of mussels, and the quantitative occurrence of planktivorous fish are important factors regulating fluxes of material through planktonic communities in coastal marine environments. We suggest that these factors be made an integral part of any evaluation of the structure and functioning of coastal marine ecosystems.

1. Introduction

Controlling factors

Production and decomposition of organic material in fjords, bays, estuaries, and other coastal regions are controlled by a large number of physical, chemical, and biological factors. Tidal changes, coastal currents and changing sea levels induce the major physical changes, while important chemical effects arise from discharges from human activities on land and in the water. Exchange of nutrients between sediments and water is also an important element. Finally, the biological structure includes a variety of attached and free-living primary and secondary producers in the sediment and in the water column. To study this complex ecosystem, it is necessary to include all major pools and fluxes of organic matter.

Objectives

The main objective of this study was to describe important controlling factors for growth of bacteria and phytoplankton. An important part of the project was to evaluate new methods and to apply these techniques to analyses of natural plankton communities.

2. Material and methods

Bacterial growth

Bacterial growth was determined by means of ^{3}H-thymidine incorporation into DNA (Furhman & Azam, 1980; Riemann et al., 1982; Riemann, 1984). A conversion factor is needed to derive

Batch cultures

Bacterial biovolume

Image analysis

bacterial cell production from thymidine incorporation data. Previous examinations using cultures from various environments suggest that conversion factors may vary 10-fold. We examined the conversion factor from experiments using batch cultures of natural assemblages of coastal marine bacteria. The average conversion factor obtained from 63 growth cycles was 1.1 x 10^{18} cells mol^{-1} thymidine incorporated (SE = 0.05, Riemann et al., 1987), which is close to the original theoretical conversion factor (Fuhrman & Azam, 1980).

Bacterial biovolume production is estimated by multiplying cell production by the mean cell volume of the population. Traditionally, the latter has been measured manually using epifluorescense microscopy; a tedious technique which produces very uncertain results. We used image analysis in combination with epifluorescence microscopy of acridine orange stained plankton samples. A program was developed for discrimination and binary segmentation of digitized video images, taken by an ultra sensitive video camera mounted on the microscope (Bjørnsen, 1986a). This system is capable of analyzing 20 microscope fields with approximately 400 bacteria within 10 min.

The calculated biovolume is converted to carbon by another conversion factor. This conversion factor was previously based on theoretical assumptions (Bjørnsen & Riemann, 1988). Direct estimates of biovolume by means of image analysis and of carbon biomass from batch cultures of natural bacterial assemblages gave an average

Carbon conversion factor

conversion factor from biovolume to carbon factor biomass of 0.35 pg C μm^{-3} (± 0.03 95% confidence limit), (Bjørnsen, 1986a) compared to normally used factors ranging 0.086-0.121 pg C μm^{-3} .

Gross carbon consumption

Finally, the net carbon biomass production is converted to gross carbon consumption by means of a theoretical growth yield. This conversion factor was derived from carbon mass balances in continuous cultures of natural bacterial assemblages. Production of CO_2 and of particulate organic matter were measured by infrared gas analysis, and growth yield was calculated as net production divided by the sum of respiration and net production (Bjørnsen 1986b). An average growth yield of 20.7% (± 1.1 95% confidence limits) was obtained; a value considerably lower than the generally assumed growth yield of 60% obtained from uptake of radiolabelled substrates.

CO_2

Growth yield

Flagellate grazing on bacteria

Grazing on natural bacterioplankton by natural assemblages of heterotrophic flagellates was estimated from 1) the decrease in bacterial cell number after inhibition of cell replication by addition of penicillin and streptomycin, 2) the decrease of radioactivity in ^3H-thymidine labelled bacteria after addition of excess concentrations of unlabelled thymidine and thymine and 3) a cell balance on unmanipulated samples based on measurements of bacterial cell production and change in cell number. See Bjørnsen (1988) for further details.

Biomass of phyto- and zooplankton

Biomass and production of phyto- and zooplankton as well as growth of mussels and planktivorous fish were measured using traditional procedures (see enclosed list of references).

Enclosure experiments

Enclosure experiments were carried out in clear cylindrical plastic enclosures (1.5 m in diameter, 3-3.5 m deep with 0.1 mm thick walls) which were fixed to a pontoon bridge. The pontoon bridge contained up to 24 enclosures per experiment. Each experiments lasted 16-25 days and water samples for determinations of biological activity and biomass were taken every 1-4 days from three depths in the enclosures. Some of the enclosures were in contact with the sediment while others were closed (fig. 1).

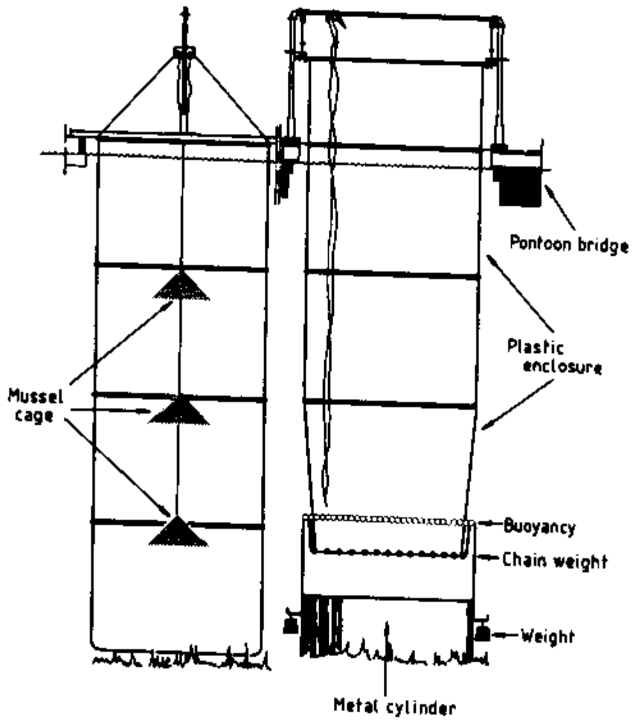

Figure 1. Closed enclosure with mussel cages added and open enclosure (open with contact to the sediment) with the buoyancy and weight system to compensate for changes in the water level. After Riemann et al. (1988).

The enclosures were used in Roskilde Fjord during April, June and September 1986 and in the Limfjord during June 1987. To compensate for wind- and tide-induced changes in the water level the enclosures connected to the sediment were constructed with a buoyancy and weight system that allowed the enclosures to withstand a change in water level of about 2 m without damage (fig. 1). Further details of the construction are found in Riemann et al., (1988).

Enclosure manipulations

The water in the enclosures were manipulated by the addition of benthic suspension feeders, planktivorous fish, nutrients, contact to the sediment and combinations of these variables (Table 1, Riemann et al., 1988; Horsted et al., 1988; Bjørnsen et al., 1988).

3. Results

3.1. Regulation of phytoplankton biomass.

Additions of mussels and nutrients

Additions of mussels and nutrients caused marked changes in the phytoplankton biomass depending on the season. Contrarily, additions of planktivorous fish revealed that zooplankton grazing at most reduced 20% of the produced phytoplankton biomass during the experimental periods (fig. 2).

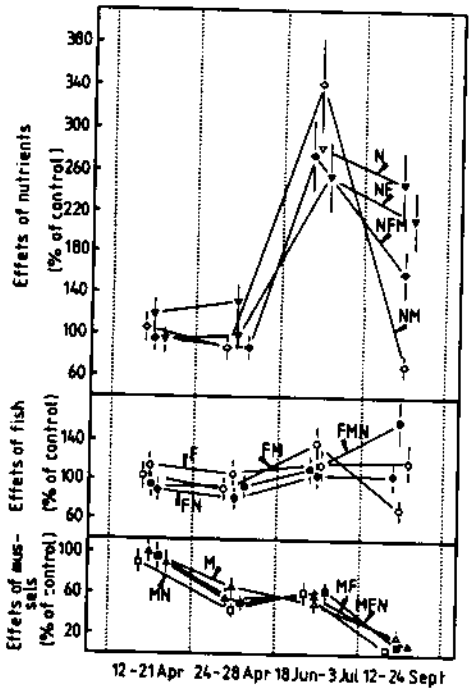

Figure 2. Effects of the addition of mussels (M), fish (F) and nutrients (N) (% of controls) on the phytoplankton biomass during April, June/July, and September 1986. Bars represent SE. After Riemann et al. (1988).

Effects of mussels on the phytoplankton biomass

Additions of mussels reduced phytoplankton biomass during summer and autumn to between 10 and 59% of control enclosures without mussels added, whereas no effects of mussels were found when the temperature was below about 5°C. The differences in the response pattern of mussels on phytoplankton biomass during the summer and autumn periods was caused by changes in the size distribution of the phytoplankton biomass.

Picoplankton

At the start of the experiment in June 1986, the number of picoplankton cells was 0.4×10^5 ml^{-1}, corresponding to 6.5% of the total phytoplankton biovolume. The mussels induced an increase in the number of picoplankton cells and at the end of the experiment the number of picoplankton cells was $3.9-6.2 \times 10^5$ ml^{-1} in enclosures with added mussels and $0.2-4.8 \times 10^5$ ml^{-1} in controls (fig. 3a), and the picoplankton comprised 70-93% of the total phytoplankton biovolume in enclosures with added mussels compared to 4-20% in controls (fig. 3b).

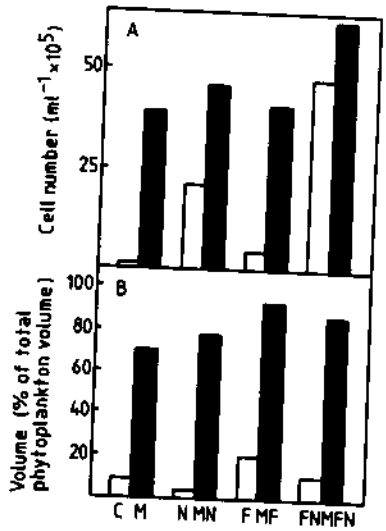

Figure 3. (A) Numbers of autotrophic picoplankton (1-2 μm) and (B) cell volume (% of phytoplankton volume) in enclosures with and without addition of mussels in June/July. C: control, M, F, N: additions of mussels, fish and nutrients, respectively. After Riemann et al. (1988).

3.2. The importance of bacteria and heterotrophic flagellates.

Oscillations between bacteria and flagellates

It has been demonstrated that heterotrophic nanoflagellates in marine environments are important as grazers of bacteria sized particles. We examined how the coupled oscillations in numbers of bacteria and flagellates were affected from the "bottom" by the rate and stability of supply of substrates for bacteria and from the "top" through predation. Tightly coupled oscillations in biomasses of bacteria and heterotrophic flagellates were found, particularly during summer. Enrichment of the system by additions of nutrients increased the flow rates (production and grazing loss of bacteria), while predator control and reduction of substrate supply (additions of mussels) reduced the flow rates (Fig. 4).

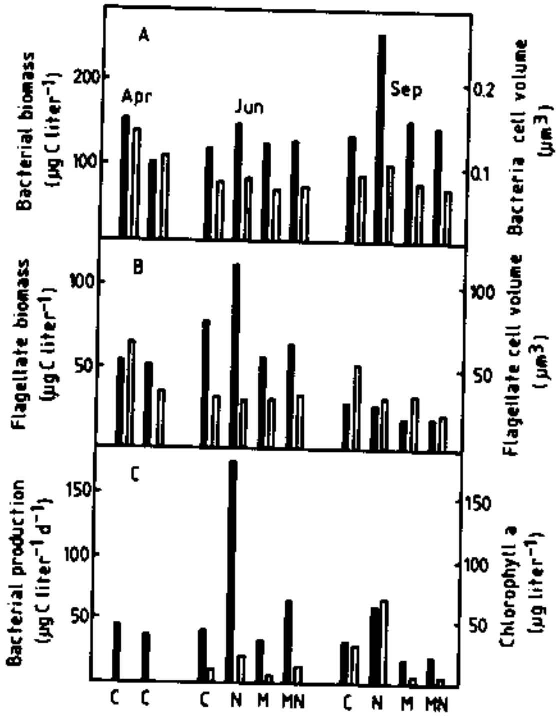

Figure 4. Effects of manipulations with nutrients and mussels during enclosure experiments in April, June, and September. a. Averages of bacterioplankton biomass (closed) and bacterial cell volume (open). b. Averages of flagellate biomass (closed) and cell volume (open). c. Averages of bacterioplankton net production (closed) and chl. a (open). April: control enclosure during two periods (0-10 d and 15-25 d from start); June and September: control enclosure (C), nutrient loaded (N), with mussels (M), and nutrient loaded with mussels (MN). After Bjørnsen et al. (1988).

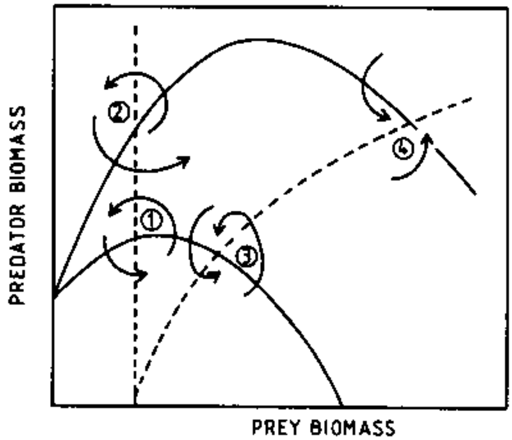

Figure 5. Theoretical effects of enrichment and top predation control on a prey-predator system. Solid curves show the effect of enrichment on the prey isocline; broken curves show the effect of top predator control on the predator isocline. Equilibrium point 1 is neutrally stable, point 2 (enrichment, no predator control) is unstable, and points 3 and 4 are stable. After Bjørnsen et al. (1988).

Ciliates

During another enclosure experiment in The Limjord, the role of planktonic ciliates were included and a carbon budget was established from daily measurements of phytoplankton and bacterioplankton and production and biomasses

This indicates that enrichment is absorbed by the system and that the increased flux of material is passed on to higher trophic levels or respired. Moreover, enrichment increased the amplitude of the coupled oscillations, while additions of mussels damped the oscillations. These observations are consistent with a simple Lotka-Volterra model demonstrating effects of enrichment and top predation control on a prey-predator system (Fig. 5).

Amplitude of oscillations

of phytoplankton, bacteria, flagellates, ciliates and meso-zooplankton. A tight coupling was found between nutrient additions and subsequent increases in phytoplankton biomass and production rates. The corresponding heterotrophic response included increased production of bacteria and biomass of ciliates (Riemann et al., submitted).

Importance of cilliates

The importance of the ciliates in the system is illustrated from the fact, that calcuated ciliate net production was almost the same as the meso-zooplankton food consumption in enclosures without nutrients but 2-3 times higher than meso-zooplankton food consumption in enclosures added nutrients. No co-occurring increases were observed in the meso-zooplankton during the experiment.

3.3. Regulation of zooplankton.

Effects of mussels, fish and nutrients on zooplankton

In the enclosure experiments, we studied effects of mussels, fish, nutrients and combinations of these on zooplankton > 45 μm. Additions mussels reduced the number of tintinnid ciliates (Tintinnopsis sp) and rotifers (Synchaeta spp.) compared to changes in control enclosures (Fig. 6).

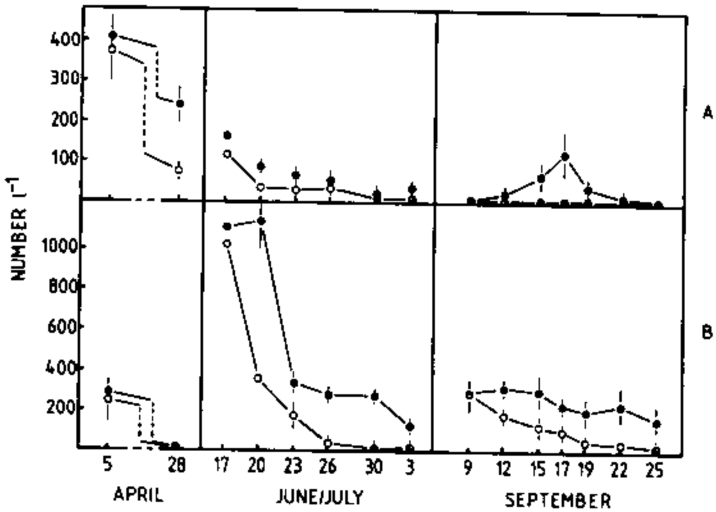

Figure 6. Numbers of (A) tintinnid ciliates Tintinnopsis spp. and (B) rotifers Synchaeta spp. during the 3 experimental periods in enclosures with added mussels (o), and without mussels (o). Bars represent standard deviation (n=12). After Horsted et al. (1988).

The average mean sizes of ciliates and rotifers were larger in enclosures added mussels, suggesting that mussels may act as a major controlling factor for small zooplankton species.

Zooplankton: dominating species

The zooplankton biomass in Roskilde Fjord is is dominated of Acartia tonsa. In contrast to the effects on small-sized zooplankton, mussels and nutrients had no effects on the abundances of the copepod A. tonsa (Horsted et al. 1988). Fish controlled the occurrence of A. tonsa, particularly during summer.

Regulation of zooplankton

Regulation of the zooplankton community is carried out from the top of the size spectrum by planktivorous fish and from the bottom of the size spectrum by mussels.

4. Discussion and conclusion

This project attached the greatest importance to methodological calibrations and validations and to quantitative descriptions of pools and fluxes of organic matter between phytoplankton, bacteria, heterotrophic flagellates, ciliates and meso-zooplankton in coastal waters. Part of the main objective was to quantify the carbon metabolism in relation to addition of nutrients, filtration by suspension-feeding bivalves and predation by planktivorous fish.

Size structure of phytoplankton biomass

The size structure of the phytoplankton biomass affects the potential of benthic suspension feeders in regulating phytoplankton biomass. Moreover, this apparently also influences the coupled oscillations in biomasses of bacteria and heterotrophic nanoflagellates. When small-sized nanophytoplankton (< 20 μm > 2 μm) dominated the biomass, small ciliates, grazing on both nanophytoplankton, flagellates and bacteria, occurred in large numbers, and changed the coupled oscillations between algae, bacteria and flagellates towards direct oscillations between algae and ciliates. Additions of nutrients strengthened the succession from flagellates towards ciliates in the oscillations and increased the amplitude of the oscillations.

Addition of mussels

Addition of mussels generally damped oscilla - tions without changing the succession of organisms involved in the oscillations. When large-sized phytoplankton (net-phytoplankton > 20 μm) dominated the biomass, coupled oscillations between algae, bacteria and flagellates occurred. Since net-phytoplankton cannot be eaten by small ciliates, food limitations prevent that they reach similar importance as found during periods where the size structure of the phytoplankton biomass is dominated by small nano-sized organisms.

A number of provisionally conclusions also point out intimate interactions between growth and biomass of bacteria, the biomass and the size structure of phytoplankton, additions of nutrients and the physiological state of the mussels. Effects of nutrient enrichment on bacteria and heterotrophic flagellates included a greater flow through their biomasses, however, the size of their biomass remained the same. The increased flow through these microheterotrophs caused decreased stability of the system. The meso-zooplankton is mainly regulated by predation from fish and effects of filtration by mussels. The importance of food quality and quantity for zooplankton is not well described, but it is clear that fish predation mainly regulate large size-fractions of meso-zooplankton while mussels mostly filtrate small size-fractions.

Decreased stability

Carbon budget

Our analyses of the carbon budget are based on qualitative and quantitative examinations of pools and fluxes. A large number of assumptions are made and the whole approach is far from perfect. Nevertheless, major trends have been identified and evaluated in an ecosystem context. Such analyses are necessary before comprehensive models can be made on the cycling of organic matter in coastal marine ecosystems.

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Part 1 of the list includes references produced only by the participants of the project. Part 2 includes other cited literature.

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for Fisheries and Marine Research, Charlottenlund Castle,
DK-2920 Charlottenlund

Abstract:
The project represents the initial stage of an overall evaluation of the flux of organic matter in coastal waters. From the results of a number of scenarios of the carbon budget appears that the balance between nutrient additions, the size distribution of phytoplankton, the physiological conditions of mussels, and the quantitative occurrence of fish are important factors regulating the structure and functioning of coastal marine ecosystems.

Terms:
production; decomposition; eutrophication; nutrients; coastal waters; bacteria: phytoplankton; analytical methods

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Resumè:
Projektet repræsenterer et første trin i en samlet evaluering af omsætningen af organisk stof i de kystnære områder. Resultaterne fra en række kulstofbudgetter viste, at balancen mellem tilsætning af næringsstoffer, planteplanktonets størrelses-fordeling, muslingernes fysiologiske tilstand og forekomst af fisk er afgørende for de kystnære, marine økosystemers struktur og funktion.

Emneord:
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