# DOES THE RED TIDE-FORMING DINOFLAGELLATE *NOCTILUCA SCINTILLANS* FEED ON BACTERIA?

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SARSIA

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Investigations were made to determine if *Noctiluca scintillans* is able to remove and ingest bacteria and thus influence bacterial numbers in the water column, before and after a *Noctiluca* 'bloom' (red tide). Laboratory experiments were performed with bacteria-sized fluorescent latex microspheres of 0.5, 1.1, and 4.5 µm diameter, as well as with live cells of the bacteria *Vibrio* sp. and *Serratia_plymuthica*. Results indicate that *Noctiluca* can remove microspheres and bacteria at a rate of $10^4$ to $10^6$ individual$^{-1}$ hour$^{-1}$, under the given laboratory conditions. Data on abundance of *Noctiluca*, different groups of bacteria, yeasts, nanoflagellates, and microbial exoenzyme activity in the southern North Sea are compared. Although removal of bacteria by *Noctiluca* in the field is generally not reflected in the occurrence of these organisms, it is possible that *Noctiluca* ingests bacteria in the winter when other food sources are scarce. During its exponential growth, *Noctiluca* may remove bacteria from the water column together with other potential and actual food organisms trapped in its mucoid webs.

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## INTRODUCTION

Every year, especially in the summer months June-July, a mass development of the heterotrophic dinoflagellate *Noctiluca scintillans* MACARTNEY, 1810 takes place in the southern North Sea, resulting in a red tide (UHLIG & SAHLING 1990). The maximal abundance of viable cells in the mixed water column may average between 100 and 800 cells litre$^{-1}$, but far higher numbers may occur in the surface waters (UHLIG 1990). A breakdown of the 'bloom' always occurs in August.

Studies on the ecology of *Noctiluca* have shown no evidence of food preference (UHLIG & SAHLING 1990). On the contrary, voracious *Noctiluca* cells may ingest, in addition to other phytoplankton, smaller cells of their own species, fish eggs and 'micro-copepods' (ENOMOTO 1956), copepod eggs (SEKIGUCHI & KATO 1976; KIMOR 1979; DAAN 1987), *Artemia* eggs, cellulose, egg yolk, and indigestible particles such as glass or chalk (UHLIG 1972). Besides raptorial feeding, *Noctiluca* cells are also capable of producing mucoid webs, in which small diatoms and other algae or detritus are trapped and subsequently ingested (UHLIG 1983). Although phytoplankton is the main source of food for *Noctiluca*, HANSLIK (1987) states that the available biomass of phytoplankton is not sufficient to support the mass development of *Noctiluca* in the southern North Sea, and that the total seston must also be considered as potential food component. The possibility that bacteria may serve as food source for starving *Noctiluca* cells was not excluded. Bacteria could play a role in the diet of *Noctiluca*, with a possible impact on bacterial numbers in the water column (W. Greve, pers. commn).

The purpose of this paper is to investigate bacterial uptake by *Noctiluca* in laboratory experiments with suitably sized fluorescent microspheres as well as with living bacteria. Comparisons of field data for *Noctiluca* occurrence with that of bacteria and other unicellular organisms at the island Helgoland are also presented and discussed.

## MATERIAL AND METHODS

### *Laboratory experiments*

Suspensions of the unicellular green alga *Dunaliella tertiolecta* (Butcher) were maintained as food for *Noctiluca*. The algae were cultivated in autoclaved, filtered seawater containing $NaNO_3$ and $Na_2HPO_4$ (final concentration 75 mg litre$^{-1}$ and 5 mg litre$^{-1}$, respectively) according to Guillard & Ryther (1962, modified), and enriched with soil extract (Erdschreiber). *Noctiluca* was cultivated in autoclaved, filtered seawater. Cultures were fed at intervals of 2-3 days with a suspension of healthy *Dunaliella* (i.e. in exponential growth phase) as needed, so no food deficit occurred. Well-fed *Noctiluca* cells are characterized by compact food vacuoles and strings of *Dunaliella* adhering to the tentacles (Uhlig 1983, Uhlig & Sahling 1995). Seawater was renewed every 5 days; *Noctiluca* was retained on a 200 µm gauze sieve and transferred to fresh medium. *Noctiluca* and its algal food were kept at room temperature (about 20° C); illumination was provided by natural light from an east window.

The bacteria *Vibrio* sp. (List-7) and *Serratia plymuthica* (strain 19 red) were originally isolated from the North Sea and maintained on agar slants with ZoBell 2216E medium. For use in feeding experiments, fresh bacteria material was removed from the agar and suspended in autoclaved seawater. The concentrations of bacteria were determined, after suitable dilution, by the spread plate method.

Fluorescent latex microspheres with average diameters of 0.5, 1.1, and 4.5 µm were obtained from Polysciences Ltd., Eppelheim, Germany (Fluoresbrite carboxylate yellow-green microspheres). The concentration in the batches was determined by serial dilution with filtered seawater, filtration onto a Sartorius 0.45 µm membrane filter rinsed with a 10 % solution of Tween 80 to prevent clumping (Nygaard & al. 1988) and subsequent examination with epifluorescence microscopy. Since clumping did not occur to any extent in the batches received, no pre-treatment for the experiments was necessary. Laboratory experiments were performed in covered, round glass evaporating dishes, at room temperature (20° C). Prior to the experiments, *Noctiluca* cells were starved 24-48 hours. For microscopical examination of microspheres in the food vacuoles (Zeiss standard, 16 x and 40 x objectives), about 10 *Noctiluca* were added to 20 ml autoclaved, filtered seawater. For measurements of microsphere and bacteria removal rates, larger numbers of *Noctiluca* were required (120 *Noctiluca* in 40 ml seawater and 40 in 20 ml per vessel, respectively). The final concentrations of microspheres used were $10^4$-$10^8$ ml$^{-1}$, resembling concentrations of bacteria in the North Sea at Helgoland (Rieper-Kirchner 1989).

Turbidity of bacteria and microsphere suspension was measured in 1 cm optical glass cuvettes on an Eppendorf analog photometer 1101 M with a 400-600 nm filter. From a calibration curve prepared according to Drews (1968), the particle numbers were determined from the equation

$$\text{particles ml}^{-1} = 3.686 \cdot 10^8 - (1.879 \cdot 10^8 \ \log T\ (\%))$$

with $r^2 = 0.995$ and where $T$ is the measured transmission. Photographs were taken on 200 ISO film for color and 100 ISO for black and white prints, using a Zeiss standard epifluorescence microscope.

### *Field data*

The study area was the Helgoland Roads between the main and dune islands, station 'Kabeltonne' (cable buoy). The methods of sampling and counting *Noctiluca* have been described by Uhlig & Sahling (1990). Long-term studies on the abundance of bacteria in the same area have been carried out by Gunkel & Klings (1962-1993) using the plate method and since 1987 by direct counts with acridine orange stain. The latter method also includes nanoflagellates. Specialized bacteria capable of degrading mannitol, alginate, starch, agar and cellulose were determined on selective media (Rieper-Kirchner 1989). *Escherichia coli* and coliform bacteria were counted on Endo nutrient pads moistened with seawater (Sartorius GmbH, Germany). Up to 1992, yeasts were counted according to Meyers & al. (1967) and from 1993 onwards on Sartorius Wort nutrient pads. *Noctiluca*, bacteria, nanoflagellates and yeast numbers were counted at weekly intervals. Exoenzyme activity was determined with methylumbelliferyl substrates according to Obst & Holzapfel-Pschorn (1988) and Kirchner (1995). Measurements were made with a spectrofluorimeter (Jobin Yvon JY3, France) at 364 nm excitation and 445 nm emission.

## RESULTS

### *Laboratory experiments with fluorescent microspheres*

Table 1 presents the results of experiments with *Noctiluca* and fluorescent microspheres. Each experiment lasted 1-24 hours, after which *Noctiluca* cells were immediately examined under the microscope. The living cells were not crushed or harmed in any way, so that the microspheres observed were actually within the intact food vacuoles. Due to the small size of microspheres (0.5-4.5 µm) in relation to *Noctiluca* (on average 0.3-0.8 mm), densities of more than 20 microspheres in the food vacuoles were

only estimated, and the higher values in Table 1 do not represent exact counts. The total numbers of microspheres from all vacuoles (generally 1-5 food vacuoles per individual were seen) were used in the calculations.

Large fluctuations were found in the numbers of microspheres taken up by *Noctiluca*. For example, some individuals ingested 4.5 µm spheres after only 1 hour, while others showed no uptake after 4 hours, with concentrations of $2 \cdot 10^5$-$2 \cdot 10^6$ microspheres $ml^{-1}$. The number of spheres in the vacuoles varied from 0 to over 100, among *Noctiluca* in the same experimental vessel.

For the smaller microspheres 1.1 and 0.5 µm, the duration of exposure was 3.5-24 hours. Here again, there were differences in uptake among individual *Noctiluca* which cannot be explained by differences in microsphere concentrations. An exception occurred in one vessel, where microspheres were observed in the vacuoles of all *Noctiluca* present, namely those of 0.5 µm diameter at the highest concentration offered, $4.3 \cdot 10^6$ $ml^{-1}$.

To determine the removal rate of microspheres from a suspension by means of *Noctiluca*'s mucoid webs (i.e. predation on the particles by mean of the web), turbidity measurements were made at the start and end of an experiment, and the results compared to a calibration curve made for 1.1 µm spheres. The turbidity, measured as change in percent light transmission, was recorded at time zero, and after 6 and 24 hours. For each of four different microsphere concentrations, 120 *Noctiluca* in vessels with 40 ml seawater were used. Table 2 shows the results. The calculated uptake rate does not mean all these spheres were actually ingested, but represents those that were effectively removed from the suspension by entrapment in the mucoid webs. Before each measurement was made, the mucoid webs and spheres therein were carefully removed with a Pasteur pipette and the vessels stirred. This was necessary to reduce errors, since some of the spheres settled out to the bottom and sides of the vessels, particularly after 24 hours and with the most concentrated suspensions. Results were calculated assuming survival and uniform activity of all 120 *Noctiluca* introduced at time zero. After 24 hours, however, the recovery of *Noctiluca* was 110, 101, 130, and 123 individuals in vessels 1 to 4, respectively.

Table 2 shows that an increase in percent light transmission, corresponding to a decrease in suspension density, was measurable in all four vessels after 6 hours. A further density decrease occurred after a total of 24 hours in all vessels except No. 2; here the increase may be due to experimental error. The calculated removal of spheres from the suspension shows generally a greater removal

Table 1. *Noctiluca scintillans*: Uptake of 4.5, 1.1, and 0.5 µm fluorescent microspheres; volume = 20 ml per vessel.

| Diameter of microspheres | Concentration of microspheres ($ml^{-1}$) | Exp. No | Duration of Experiment ( hour ) | Number of *Noctiluca* per vessel (n) | Uptake of microspheres Range | Mean | Median |
|---|---|---|---|---|---|---|---|
| 4.5 µm | $2.0 \cdot 10^5$ | 2 / 94 | 1 | 10 | 0-8 | 1.3 | 0 |
| | | 2 / 94 | 2 | 9 | 0 | 0 | 0 |
| | | 2 / 94 | 4 | 10 | 0-20 | 2.7 | 0 |
| | | 1 / 93 | 4 | 10 | 0-110 | 26.5 | 0 |
| | | 1 / 94 | 4 | 10 | 0-100 | 15.8 | 0 |
| | $2.0 \cdot 10^6$ | 2 / 94 | 1 | 11 | 0-21 | 2.7 | 0 |
| | | 2 / 94 | 2 | 10 | 0-15 | 2.7 | 0 |
| | | 2 / 94 | 4 | 10 | 0-20 | 2.0 | 0 |
| | | 1 / 93 | 4 | 11 | 0- >140 | 28.4 | 11 |
| | | 1 / 94 | 4 | 7(3 lost) | 0-20 | 6.9 | 0 |
| 1.1 µm | $2.5 \cdot 10^4$ | 3 / 94 | 3.5 | 10 | 0 | 0 | 0 |
| | | 3 / 94 | 24 | 10 | 0 | 0 | 0 |
| | | 4 / 94 | 24 | 8 | 0- >200 | 60.5 | 29.5 |
| | $2.5 \cdot 10^5$ | 3 / 94 | 24 | 11 | 0-200 | 34.7 | 8 |
| | | 4 / 94 | 18.5 | 10 | 0-500 | 103.5 | 15 |
| | $2.5 \cdot 10^6$ | 3 / 94 | 24 | 10 | 0-300 | 61.3 | 0 |
| | | 4 / 94 | 18 | 10 | 0-500 | 103.8 | 40 |
| 0.5 µm | $4.3 \cdot 10^4$ | 7 / 94 | 22.5 | 11 | 0-50 | 16.5 | 3 |
| | $4.3 \cdot 10^5$ | 7 / 94 | 22.5 | 12 | 0-100 | 18.3 | 7.5 |
| | $4.3 \cdot 10^6$ | 7 / 94 | 22.5 | 10 | 40-250 | 143 | 135 |

rate by *Noctiluca* in vessels with higher microsphere concentrations. A more rapid removal by *Noctiluca* individuals took place during the first 6 hours, than over the 24 hours taken as a whole.

Fig. 1 shows the accumulation of the yellow-green fluorescent spheres in the food vacuoles, typically tube-shaped or spherical, in active *Noctiluca* cells from vessels No. 2 and 3 as described in Table 2 (photographs A and B, respectively). Fig. 2 shows the form of a mucoid web with the microspheres entrapped therein, demonstrating in miniature a remarkable resemblance to meter-long parabolic mucoid strings of *Noctiluca* observed in the water column by divers in the field (Omori & Hamner 1982).

*Laboratory experiments with live bacteria*

In order to determine the quantitative removal of live bacteria from a suspension by *Noctiluca*, two experiments were performed with *Vibrio* sp. and *Serratia plymuthica*. In the first, bacterial concentrations were determined by plate counts, since both species form characteristic colony forming units (CFU), easily distinguishable from background bacteria accompanying *Noctiluca* cultures. The duration of the first experiment was 22 hours, n = 40 *Noctiluca* for each treatment. The results, presented in Table 3, indicate a removal of $10^4$-$10^5$ *Vibrio* sp. per individual *Noctiluca* per hour. For *Serratia*, on the other hand, an increase in the bacterial numbers occurred, even in the absence of added nutrients, which may have masked a possible uptake by *Noctiluca*. The growth of *Serratia* may have been favored by *Noctiluca* mucus production.

The second experiment was performed with higher initial concentrations of bacteria; changes in the density of the suspensions were determined photometrically. Controls were set up to determine the change in bacterial density without *Noctiluca*. The final bacterial numbers in vessels containing *Noctiluca* (n = 40 *Noctiluca* per vessel) were subtracted from those in the controls, to determine the removal rate. The duration was 18 hours, under the same conditions as above. Table 4 shows that the removal of bacteria from the suspensions was greater at higher initial bacterial concentrations for both *Vibrio* and *Serratia*. Yet the concentrations of bacteria used here appeared to have a detrimental effect on *Noctiluca*. Although with both species, *Noctiluca* initially produced copious mucoid feeding webs in which numerous bacteria were entrapped, and thus removed from the suspensions,

Table 2. *Noctiluca scintillans*: Removal of 1.1 μm fluorescent microspheres from a seawater suspension, determined photometrically; n = 120, volume = 40 ml per vessel.

| Vessel No. | % Transmission (spheres $ml^{-1}$) at time zero | after 6 hours | after 24 hours | Removal spheres $ind.^{-1}hour^{-1}$ during first 6 hours | during 24 hours |
|---|---|---|---|---|---|
| 1 | 6.9 ($2.1 \cdot 10^8$) | 8.7 ($1.9 \cdot 10^8$) | 9.3 ($1.9 \cdot 10^8$) | $1.1 \cdot 10^6$ | $3.4 \cdot 10^5$ |
| 2 | 24.7 ($1.1 \cdot 10^8$) | 29.0 ($9.4 \cdot 10^7$) | 26.8 ($1.0 \cdot 10^8$) | $7.3 \cdot 10^5$ | $9.2 \cdot 10^4$ |
| 3 | 73.5 ($1.8 \cdot 10^7$) | 77.3 ($1.4 \cdot 10^7$) | 80.9 ($1.0 \cdot 10^7$) | $2.3 \cdot 10^5$ | $1.1 \cdot 10^5$ |
| 4 | 86.6 ($4.5 \cdot 10^6$) | 87.8 ($3.4 \cdot 10^6$) | 91.0 ($4.9 \cdot 10^5$) | $6.2 \cdot 10^4$ | $5.6 \cdot 10^4$ |

Table 3. *Noctiluca scintillans*: Removal of live bacteria *Vibrio* sp. and *Serratia plymuthica* from a seawater suspension, as determined by the plate method; n = 40, volume = 20 ml per vessel.

| Bacteria species | cfu $ml^{-1}$ at time zero | cfu $ml^{-1}$ after 22 hours | Removal of cfu $ind.^{-1}$ $hour^{-1}$ |
|---|---|---|---|
| *Vibrio* sp. | $3.44 \cdot 10^7$ | $2.28 \cdot 10^7$ | $2.64 \cdot 10^5$ |
| *Vibrio* sp. | $3.31 \cdot 10^6$ | $2.00 \cdot 10^6$ | $2.98 \cdot 10^4$ |
| *Serratia plymuthica* | $8.59 \cdot 10^6$ | $2.06 \cdot 10^7$ | – |
| *Serratia plymuthica* | $8.51 \cdot 10^5$ | $1.64 \cdot 10^6$ | – |

Fig. 1 (next page). *Noctiluca scintillans*: Fluorescent microspheres (1.10 □m) in the food vacuoles; concentrations in the surrounding medium (arrow) were $9.55 \cdot 10^7$ spheres $ml^{-1}$ (A) and $1.32 \cdot 10^7$ $ml^{-1}$ (B) 160 x.

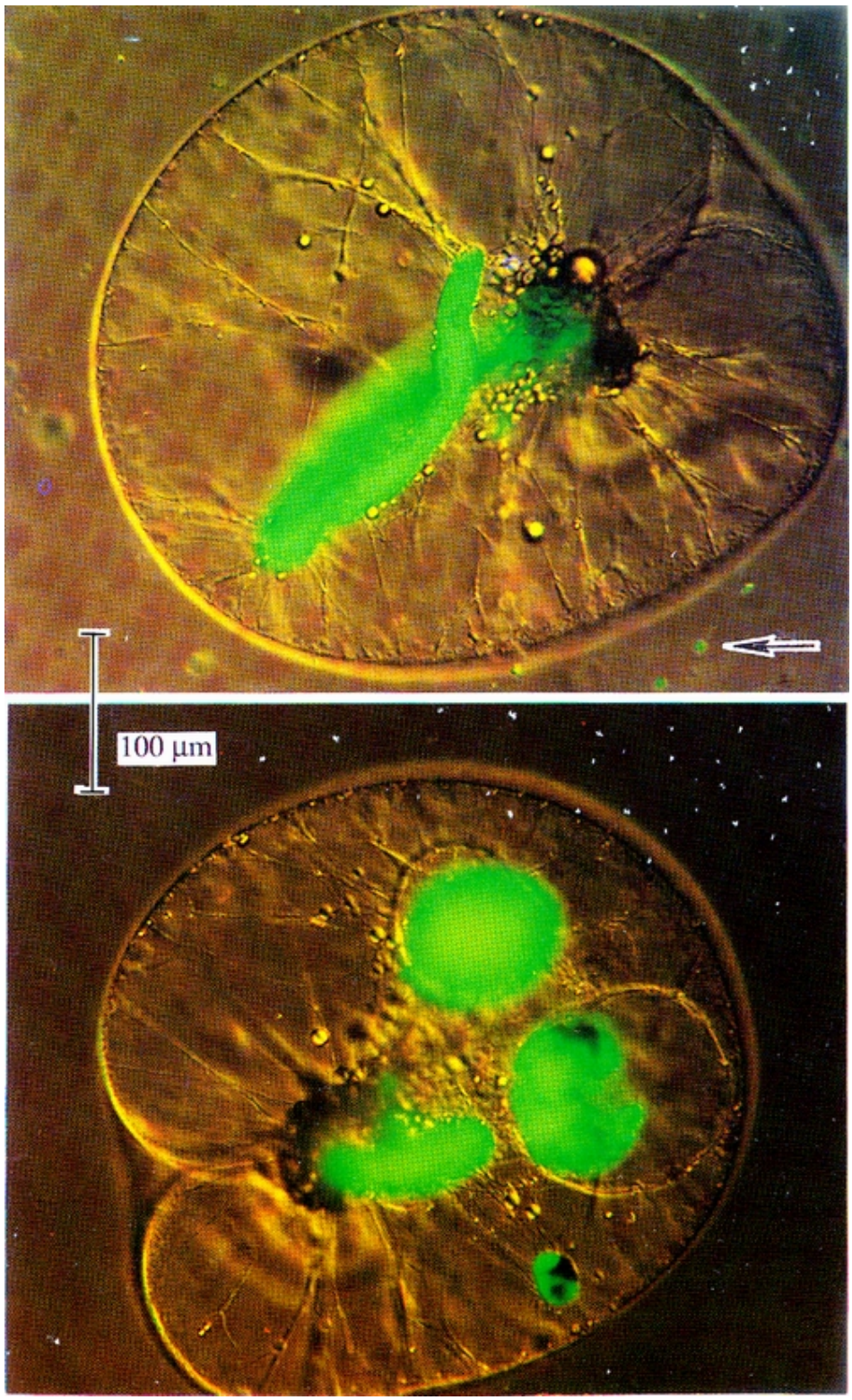
100 µm

Fig. 2. *Noctiluca scintillans*: Mucoid feeding web with fluorescent microspheres entrapped therein. 160x.

the *Noctiluca* cells became sluggish and ceased feeding movements. Those which had received *Vibrio* showed a rapid recovery, however, when transferred immediately to fresh seawater, and normal tentacle movements resumed. *Serratia*, on the other hand, appeared to inactivate *Noctiluca* to a greater extent at the concentrations in this experiment, although these were lower than those for *Vibrio*. About one fourth of the cells with *Serratia* were irreversibly damaged, the tentacles had vanished, there were pink oily globules in the cytoplasm but no recognizable vacuoles or other structures. These cells soon burst, leaving the globules on the surface on the water. Most of the remaining cells recovered in fresh seawater, and began feeding on the *Dunaliella* offered. The lower concentrations of *Serratia* in the previous experiment (Table 3) did not appear to harm *Noctiluca*. The higher concentrations, however, apparently had a poisonous effect, possibly due to *Serratia*'s pigment or enzymes and other metabolic products which accumulated in the medium.

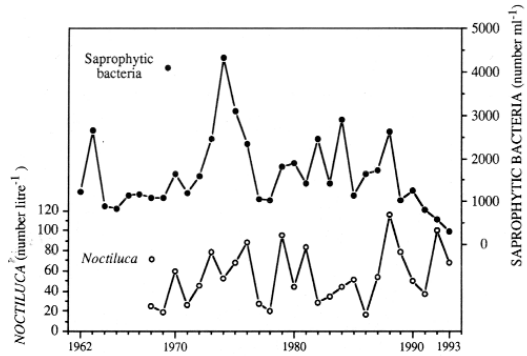


Fig. 3. *Noctiluca scintillans*: Mean seasonal abundance at Helgoland Roads from 1968 to 1993 and mean annual abundance of saprophytic bacteria at Helgoland Roads from 1962 to 1993.

*Field data*

The seasonal occurrence of *Noctiluca* at the Helgoland Roads has been studied continuously for more than 20 years (UHLIG & SAHLING 1990, 1995). Results show that the buildup of a 'bloom' may begin as early as April or as late as June. Maximum abundance is reached in June-July; an abrupt breakdown of the summer population always takes place in August. Fig. 3 shows the mean seasonal abundance from 1968 to 1993.

At the same station, the occurrence of saprophytic bacteria has been investigated by GUNKEL & KLINGS (1962-1993). Saprophytes are capable of a rapid uptake of easily degradable substrates and thus may be considered as indicators of intensive organic breakdown processes and as indicators of pollution (RHEINHEIMER 1991). Characteristic for the annual curve of mean monthly values (GUNKEL & KLINGS 1962-1993) is a small peak in February-March, then a sharp increase, often resulting in a large, single-peak maximum in June or July. Minimum values occur in the winter months. Fig. 3 shows the mean annual abundance of saprophytic bacteria covering a period of 30 years. From 1989 to 1993, a more or less steady decrease in saprophyte numbers has taken place. The mean annual values for 1992 and 1993 were the lowest since measurements were begun in 1962.

Direct bacteria counts (AODC) made from the same water samples are, as generally reported in the literature, much higher than those obtained with the culture method. Direct counts include, however, not only active but also inactive and dead bacteria cells, as well as those

Table 4. *Noctiluca scintillans*: Removal of live bacteria *Vibrio* sp. and *Serratia plymuthica* from a seawater suspension, determined photometrically; n = 40, volume = 20 ml per vessel.

| Bacteria species | Bacteria $ml^{-1}$ at time zero | Bacteria $ml^{-1}$ after 18 hours | Removal of bact. $ind.^{-1}$ $hour^{-1}$ |
|---|---|---|---|
| *Vibrio* sp. alone | $3.50 \cdot 10^8$ | $3.18 \cdot 10^8$ | – |
| *Vibrio* sp. + *Noctiluca* | $3.50 \cdot 10^8$ | $2.73 \cdot 10^8$ | $1.25 \cdot 10^6$ |
| *Vibrio* sp. alone | $1.80 \cdot 10^8$ | $1.63 \cdot 10^8$ | – |
| *Vibrio* sp. + *Noctiluca* | $1.80 \cdot 10^8$ | $1.36 \cdot 10^8$ | $7.50 \cdot 10^5$ |
| *Serratia plym.* alone | $8.90 \cdot 10^7$ | $1.07 \cdot 10^8$ | – |
| *Serratia plym.* + *Noctiluca* | $8.90 \cdot 10^7$ | $9.12 \cdot 10^7$ | $4.39 \cdot 10^5$ |
| *Serratia plym.* alone | $4.40 \cdot 10^7$ | $5.63 \cdot 10^7$ | – |
| *Serratia plym.* + *Noctiluca* | $4.40 \cdot 10^7$ | $5.04 \cdot 10^7$ | $1.64 \cdot 10^5$ |

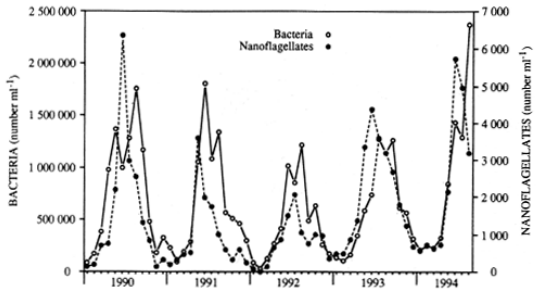


Fig. 4. Direct counts of bacteria and nanoflagellates (AODC): Mean monthly values at Helgoland Roads 1990-1994.

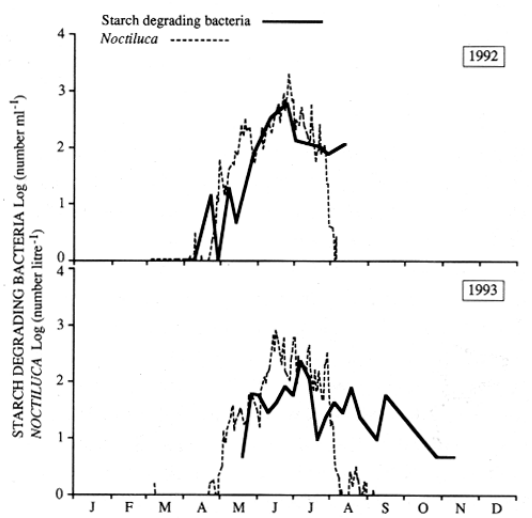


Fig. 5. *Noctiluca scintillans* abundance vs starch degrading bacteria at Helgoland Roads: Average monthly values for 1992 and 1993.

which require culture methods other than with standard 2216E agar. Fig. 4 presents recent results together with nanoflagellate data (flagellates 2-20 µm). After a minimum in January-February, there is generally a steady increase to a broad maximum extending from May to August/September. In contrast to the saprophyte numbers, no decrease has been found with AODC values during the last few years. On the contrary, the maxima have remained constant at about $10^6$ bacteria $ml^{-1}$. Nanoflagellate numbers range from $10^2$-$10^4$ $ml^{-1}$. The summer maximum is represented by a single peak, occurring about one month before the bacteria maximum.

Culture methods were employed not only for the determination of the overall numbers of saprophytic bacteria, but also to distinguish between different groups of microorganisms. With selective media, bacterial degraders of ecologically important substrates such as mannitol, alginate, cellulose, starch and agar were determined. The period of investigation was concentrated on the months during and after a *Noctiluca* bloom, from 1991 to 1993. The results show that the abundance curves for starch, cellulose and alginate degrading bacteria generally ran parallel to that for *Noctiluca*, particularly in 1992-1993. This means that an increase of these bacteria groups occurred during the active growth and feeding phase of *Noctiluca*, during the buildup of a 'bloom'. Fig. 5 shows a representative example with *Noctiluca* and starch degraders. No correlation is apparent between *Noctiluca* abundance and agar or mannitol degrading bacteria. A noted decrease in the numbers of specialized bacteria, corresponding to an increase in *Noctiluca* numbers, was not recognizable during the period of investigation.

Yeast values have been determined at the Helgoland Roads nearly continuously from 1964 to the present. Average monthly values range from 100 $litre^{-1}$ to a record

Table 5. Exoenzyme activity at Helgoland Roads May-August 1992: substrate turnover nMol $hour^{-1}$ at 18° C.

| Date | MUF-ß-D-glucoside | MUF-ß-D-cellobio-pyranoside | MUF-N-Acetyl-ß-D-glucosaminide |
|---|---|---|---|
| 7 May | 0.0071 | 0.0019 | 0.0072 |
| 27 May | 0.0118 | 0.0040 | 0.0081 |
| 10 June | 0.0382 | 0.0067 | 0.1202 |
| 17 June | 0.0282 | 0.0062 | 0.0179 |
| 1 July | 0.0307 | 0.0044 | 0.0203 |
| 29 July | 0.0299 | 0.0058 | 0.0216 |
| 12 August | 0.0265 | 0.0065 | 0.0254 |
| 26 August | 0.0145 | 0.0036 | 0.0182 |

maximum in September 1984 with 9555 litre$^{-1}$. Values of 1000 litre$^{-1}$ were also found in March 1985, July 1992 and August 1993. The highest yeast numbers occurred most frequently in the summer and early autumn, from June through October.

*Escherichia coli* and coliform bacteria were determined in 1993 and 1994. In 1993 maximum values occurred in August with a monthly mean of 5140 litre$^{-1}$; in 1994 the maximum occurred in September with a mean of 4710 litre$^{-1}$. In the winter months, frequently no coliforms were found in the samples.

Exoenzyme activity, an indicator for the self-purification capability of a body of water, was determined at bimonthly intervals from May through August 1992 (Table 5). The MUF-substrates used in the analyses (MUF-ß-D-glucoside, MUF-ß-D-cellobiopyranoside and MUF-N-Acetyl-ß-D-glucosaminide) were chosen to detect glucosidase, cellulase and chitinase activity, respectively. Results show that for all three substrates, the most rapid turnover rates took place in the early June sample, indicating higher bacterial activity at this time.

## DISCUSSION

In the laboratory *Noctiluca* was able to remove live bacteria species *Vibrio* sp. and *Serratia plymuthica*, both of which are rods with a diameter of 0.5-0.8 µm and length of ca 1-2 µm, as well as inert bacteria-sized microspheres, from a seawater suspension. Whereas the bacteria, which were unlabeled, could not be distinguished after ingestion, the fluorescent microspheres were clearly visible in the food vacuoles of healthy, feeding *Noctiluca* cells. Although a comparison between living bacteria cells and inert latex particles must be made with reservation, under the given conditions the rates of removal of bacteria and microspheres ranged from $10^4$-$10^6$ individual$^{-1}$ hour$^{-1}$, depending on the initial concentrations introduced to the vessels and duration of experiment. These values cannot be considered absolute, but rather as working figures in the planning of subsequent experiments. In the uptake of microspheres, individual *Noctiluca* showed a large variability (Table 1), which may also be the case when bacteria instead of inert particles are offered as food. Although the uptake values for *Vibrio* and *Serratia* by *Noctiluca* may be underestimated, since bacteria growth was not taken into consideration, the use of a growth formula might imply an accuracy in the results which does not exist. The use of rotating bottles instead of static cultures in the experiments may also have given other results (BUSKEY 1995). The purpose of this paper was primarily to explore the possibility of bacteria as food for *Noctiluca*, since microorganisms have been given little attention as its potential food. In comparison to the results presented above, the food removal rates calculated for *Noctiluca* feeding on the live algae *Tetraselmis tetrathele* (G.S. WEST) BUTCHER, 1959 (size ca 10-20 µm) and *Gymnodinium nagasakiense* TAKAYAMA & ADACHI (syn. *G. mikimotoi* MIYAKE & KOMINAMI ex ODA (ca 30 µm) in the laboratory were $2{\cdot}10^3$ and $6{\cdot}10^2$ cells individual$^{-1}$ hour$^{-1}$, respectively (LEE & HIRAYAMA 1992). These differences may be due primarily to the greater sizes of the algae compared to bacteria or microspheres.

It is not to be expected, however, that *Noctiluca* removes microorganisms or phytoplankton at these rates from the water column in the sea; as a voracious non-selective feeder, many organisms of different sizes are likely to be either directly engulfed or trapped in the mucoid feeding webs. Moreover, uptake also depends on the extent of mucoid web production, which in turn is influenced by factors such as density of *Noctiluca* and turbulence (wave action).

Bursts of intense *Noctiluca* feeding activity might be expected to be reflected in the seasonal abundance of potential food organisms. According to UHLIG & SAHLING (1990), after existing under poor conditions in the winter, *Noctiluca* cells attain an optimal nutritional status during May-June, their main growth phase. The period of maximum *Noctiluca* abundance is usually June-July. At the end of July or early August, cells become irreversibly damaged and stop feeding, prior to breakdown of the 'bloom'. Thus, if *Noctiluca* is removing large numbers of bacteria and other organisms from the water column, this would occur in May-June during exponential growth, rather than when the cells later accumulate in surface waters and create a visible 'bloom' or red tide.

During the spring months, however, there is no marked decrease in bacterial numbers. On the contrary, saprophyte counts usually increase at this time, with a general maximum in June-July. The same applies to bacteria determined with AODC, which also show an increase in May, with high numbers extending throughout the summer. Specialized bacteria on selective media either increased concomitantly with the *Noctiluca* growth phase or showed no apparent correlation. In June 1992 the greatest exoenzyme activity, representing microbial substrate turnover, corresponded with the summer maximum of overall saprophytes, during which time *Noctiluca* numbers were still relatively high. *E. coli* and coliform bacteria, colonic flora entering the sea with sewage and wastewater, generally reach their maxima during the late summer months when the water is warm and tourism is still high. Due to their relatively low numbers at the Helgoland Roads, it is not expected that this group of bacteria is easily available as potential food for *Noctiluca*. This may not be the case in coastal waters, however, where coliform bacteria are more common and conditions are more favorable for *Noctiluca* growth.

Highest yeast numbers are generally found in the summer and early autumn, from June to October. Possible correlations between *Noctiluca* 'blooms' and the size of the yeast population have been suggested by MEYERS & al. (1967). SCHAUMANN & al. (1988) observed unusual large numbers of higher fungal propagules in connection with a *Noctiluca* red tide in the southwestern German Bight (North Sea) in July-August 1984. Although the yeast *Debaryomyces hansenii,* (ZOPF) LODDER ET KREGER-VAN RIJ which is frequently found in the North Sea (MEYERS & al. 1967), could be described as an 'ideal' food for *Noctiluca* because of ist size and tendency to remain in suspension, HANSLIK (1987) and UHLIG & al. (1995) showed that *Noctiluca* could not survive on a diet of this yeast. The accumulation of yeast cells and higher fungi during a red tide, when *Noctiluca* is no longer actively feeding, may be due to secretions and lysis of damaged cells. Not only these but also other organisms have been oberved together with high concentrations of *Noctiluca* in surface waters. The exudations from disintegrating *Noctiluca* cells would permit increased microbial production and also higher numbers of accompanying bacterivorous protozoans. The release of inorganic nutrients such as $NH_4^+$ and $PO_4^{3-}$ could also stimulate local phytoplankton production (SCHAUMANN & al. 1988). According to our observations at Helgoland, the following organisms were especially abundant with surface *Noctiluca* during recent 'blooms': jellyfish (*Aurelia*), ctenophores, diverse crustacean larvae, barnacle moults (from *Balanus*, *Elminius*), various *Ceratium* (Dinophyceae) species, as well as numerous colorless flagellates and ciliates. These associations may be more coincidental than causal, however, due to the same prevailing wind, current and temperature conditions which facilitate accumulations of *Noctiluca*. Such convergence phenomena have also been described for *Noctiluca* swarms off the western coast of Brittany (French Atlantic) by LE FÈVRE & GRALL (1970).

Attention was also given to the occurrence of nanoflagellates since, under certain conditions, heterotrophic flagellates may be major consumers of bacteria (HAAS & WEBB 1979; FENCHEL 1982). A correlation with *Noctiluca* abundance was not apparent. Further investigations are needed to determine the relations of *Noctiluca* to nanoflagellates and bacteria; as well as bacteria, nanoflagellates could become trapped in the mucoid webs and serve as potential food for *Noctiluca*.

Although the phytoplankton cycles have been studied continuously for several decades (HAGMEIER 1978; HICKEL & al. 1993), up to now no correlation has been found between a *Noctiluca* red tide and any particular diatom or other algal bloom (UHLIG 1990; UHLIG & SAHLING 1995). Predator-prey cycles with *Noctiluca* and its potential or proven food organisms have not yet been shown, although *Noctiluca* cells often contain remains of phytoplankton in the food vacuoles, and ENOMOTO (1956) states that generally 90 % of its food consists of different diatom species. This agrees with laboratory results of BUSKEY (1995), who found that the highest growth rates of *Noctiluca* were supported with diatoms, in particular *Thalassiosira* sp. HANSLIK (1987) and UHLIG & al. (1995) suggest that, since the available phytoplankton concentration is not sufficient to support the masses of *Noctiluca* which occur in the German Bight, seston - comprising plankton and detritus - should be considered as additional food source. Detritus particles or aggregates, 'marine snow', are known to harbour many microorganisms (MELCHIORRI-SANTOLINI & HOPTON in RHEINHEIMER 1991).

It is possible that a small winter population of *Noctiluca* may be existing on a diet of bacteria, whether in the water column or attached to particles, when no or little other plankton is available. During the spring exponential phase of growth, *Noctiluca* is capable of ingesting large numbers of bacteria which may be entrapped in the mucoid feeding webs, along with other food organisms, on a non-selective basis. Further investigations are needed, e.g. with labelled bacteria, to determine if and to what extent bacteria are actually removed by *Noctiluca* in the field.

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