The food removal rate by *Noctiluca scintillans* feeding on *Tetraselmis tetrathelle* and *Gymnodinium nagasakiense*.

Jung Keun Lee* and Kazutsugu Hirayama

*Noctiluca scintillans* secrets mucus around its tentacle and feeds on the materials trapped in it. The food removal rates in the suspensions of *Tetraselmis tetrathelle* and *Gymnodinium nagasakiense* were calculated using a model for filter feeder which is based on the change in food level during the feeding experiment. The clearance rate by starved *Noctiluca* varied depending upon the food level and experimental duration. With *T. tetrathelle*, the rate decreased as the food level increased over $4 \times 10^5$ cells/ml. The maximal rate in the experiments of 2-3 hour duration was about $2 \times 10^{-2}$ ml/Noctiluca/h, and that in the experiments of 4-7 hour duration was about $1 \times 10^{-3}$ ml/Noctiluca/h. With *Gymnodinium nagasakiense*, when the experimental durations were fixed at 4 or 5 hours, the rates fluctuated between $1 \times 10^{-3}$ and $7 \times 10^{-3}$ ml/Noctiluca/h throughout the food levels examined.

The food removal rate with above two kinds of algal food increased with increasing food levels up to the maximal, and, once the maximal rate was achieved, there was no further increase in spite of the increase in food level. The maximal rate was about 2000 cells (or $0.75 \times 10^{-4}$ mg Carbon)/Noctiluca/h with *T. tetrathelle* and about 600 cells (or $1.5 \times 10^{-4}$ mg Carbon)/Noctiluca/h with *G. nagasakiense*. It was supposed that *N. scintillans* might inflict considerable feeding pressure on the population growth of *G. nagasakiense* in nature.

**Key words**: the clearance rate; the food removal rate; *Noctiluca scintillans*; *Tetraselmis tetrathelle*; *Gymnodinium nagasakiense*.

*Noctiluca scintillans*, which is largely vacuolated and unarmored (200-1000 μm in size), has little motility, but the food it feeds on covers a wide range of plankton, e.g., phytoplankton, protozoa, copepods and their eggs, fish eggs, etc.¹ The voracious feeding of *Noctiluca* affects the populations of other planktonic organisms. Prasad² attributed a rapid reduction in the diatom population to the feeding of *Noctiluca*. And the feeding of *Noctiluca* on the eggs of Acartia (a copepod) and Anchovy (a small fish) significantly influenced their hatched populations.³ ⁴ In the light of these facts, the role of *Noctiluca* is important in the material transfer of plankton community.

*Noctiluca* shows a characteristic feeding mechanism, mucoidfiltration⁵: The prey is caught in a layer of mucus secreted around the tentacle and carried to cytostome by the movement of the tentacle.⁶ ⁷ The clump of algal cells on the tip of tentacle gradually grows larger with the enlargement rate depending on the food density and the volume of mucus.⁷ In my laboratory culture, the strings of mucus, several times longer than the cell length of *Noctiluca*, were often observed with algal food trapped in them. The feeding behavior in nature is the same in principle as in the laboratory culture. Omori and Hamner⁸ reported an encounter of a large number of *Noctiluca* while diving deep...
in a lagoon: The strings of mucus extended from the cells of *Noctiluca* and entangled together to from a mucus web; *Noctiluca* fed on the materials trapped in it, and, as the weight of accumulating debris eventually pulled the accumulation downward, they released themselves from the web near the bottom, some 30 meter deep, to float individually back to the surface. Thus, the food removed by mucus is not necessarily fed by *Noctiluca*.

The purpose of this study is to obtain preliminary information on the role of *Noctiluca* in the material transfer of plankton community. To serve this purpose, the clearance rates (volume swept clear) and food removal rates by the organism were obtained at various food levels using *Tetraselmis tetraethelle* and *Gymnodinium nagasakiense* as food organisms.

**Materials and Methods**

1. *Noctiluca*

*Noctiluca scintillans* was collected with a plankton net from Nomo Bay in Nagasaki Prefecture, southern Japan. It was stock-cultured on a diet of *T. tetraethelle* in about 100 ml of GF/C filtered seawater at 23°C, 22% S and on 14L: 10D light cycle. Experiments started two days after the collection and continued periodically for several months.

2. Algal food

*T. tetraethelle* and *G. nagasakiense* originated from the stocks maintained in modified Erd-Schreiber medium under the same culture conditions as in *Noctiluca* stock culture. *T. tetraethelle* for the experiment was grown in 500 ml of GF/C filtered sea water enriched with modified Erd-Schreiber medium (1:1, volumetrically). The culture was placed in the dark for several hours and the settled down cells were removed. *G. nagasakiense* was grown in 100 ml of modified Erd-Schreiber medium, and the patches formed in the culture were taken to prepare a high level of food suspension. The concentrations of these cultures were determined by averaging ten cell counts which were obtained using a haemacytometer. The food suspensions of various food levels were made by diluting the cultures with filtered sea water.

3. Estimation of carbon contents in the algal food

Hundreds ml aliquot of the food culture was passed through the fired filter pads (GF/C) every time the food removal rate experiment was conducted. These samples (7 for *T. tetraethelle* and 10 for *G. nagasakiense*) were dried up at -30°C. After being cut into pieces, they were analysed for carbon contents using a C & N corder (Yanako, Model MT 500).

4. Food removal rate experiment

The experiments were conducted at 23°C and 22% S, the most favorable temperature and salinity for the growth of *Noctiluca*. The starved *Noctiluca* were inoculated in the feeding vessels containing various levels of food suspensions, and allowed to remove the food organisms for a definite period of time. Controls without *Noctiluca* were prepared to correct for the algal growth in the feeding vessels. The food removal rates were calculated from the number of *Noctiluca*, experimental duration, volume of food medium, and the food concentrations at the start and end of the experiment.

a. Experiments with *T. tetraethelle*

*Noctiluca* were starved for 24 hours in 500 ml filtered seawater, and the aggregations formed on the surface were pipetted into a beaker containing about 100 ml of filtered sea water. Special attention was paid not to give mechanical shocks during the transfer. The *Noctiluca* suspensions were condensed again, immediately before the experiments, into as highest densities as possible (800–1000 inds./ml) by gently pipetting out the seawater beneath the aggregation. The density of *Noctiluca* was determined by averaging three cell counts of 1 ml aliquots using a chamber slide glass. One to five ml of this *Noctiluca* condensation were swiftly transferred into the experimental flasks containing 100 ml of food suspension. The higher the food concentration, the more
**Fig. 1.** Schematic diagram of the apparatus used in the feeding experiment.

*Noctiluca* were used to produce the recognizable changes in food concentration at the end of experiments. The experiments were run in continuous dim light on an apparatus designed to assure the uniform distribution of *Noctiluca* and algal food in the medium (Fig. 1). A box containing 8-10 experimental flasks was suspended over a touching bar attached alongside a horizontal rotating axis. The box was continuously shaken at a rate of six reciprocations a minute with the maximal swinging distance being about 2 cm. This shaking was confirmed to be harmless to *Noctiluca*. The durations of experiment ranged from two to seven hours.

b. Experiments with *G. nagasakiiense*

These experiments were run essentially in the same way as in the *T. tetrathelle* experiments except that they were run statically using a smaller number of *Noctiluca* in a smaller amount of food suspension. Ten to fifteen *Noctiluca* were placed in each well of Falcon multiplate (no. 3046) containing 3 ml of food suspension, and the direct light was cut off by covering a black vinyl sheet on the vessels. The experiments were run for four to five hours. The food concentrations were determined in the same way as in the experiments with *T. tetrathelle*.

5. Calculation of food removal rate

The food removal rate was calculated using a model given in Ohmori and Ikeda. The changes in food concentration in the control and feeding vessels after some period of time are respectively expressed as follows:

\[ C_t = C_0 \cdot e^{kt} \quad \text{..............(1)} \]

\[ C_{tf} = C_0 \cdot e^{(k-f)\cdot t} \quad \text{..............(2)} \]

where \( C_t \) is the final food concentration in the control (cells/ml), \( C_{tf} \) is the final food concentration in the feeding vessels (cells/ml), \( C_0 \) is the initial food concentration (cell/ml), \( t \) is the duration of experiment (hour), \( k \) is the growth coefficient (or specific growth rate) of the algal food, \( f \) is the food removal coefficient by *Noctiluca*.

Solving for \( k \) and \( f \) using equation (1) and (2),

\[ k = \frac{(\ln C_t - \ln C_0)}{t}, \]

\[ f = \frac{((\ln C_0 - \ln C_{tf})/t) + k}{t} \]

The clearance rate is calculated from,

\[ F = V \cdot f/N, \]

where \( F \) is clearance rate (ml/*Noctiluca*/h), \( V \) is the volume of food suspension (ml), \( N \) is the number of *Noctiluca* in the feeding vessels (inds.).

The mean food concentration in the feeding vessel is calculated from;

\[ C = (C_{tf} - C_0)/(k-f)t, \]

where \( C \) is the mean food concentration (cell/ml).

The food removal rate is calculated from;

\[ I = C \cdot F, \]

where \( I \) is food removal rate (cells/*Noctiluca*/h).

**Results**

Fig. 2 shows the clearance rates by starved *Noctiluca* at various food levels. The rates are expressed by dividing them into two groups on the basis of experimental duration. The cell number was converted into carbon contents using the results of chemical analyses: the average values of carbon contents in \( 1 \times 10^6 \) cells were 3.74 mg (S. D.: 0.3) for *T. tetrathelle* and 25.7 mg (S. D.: 10.17) for *G. nagasakiiense*. With *T. tetrathelle*, the clearance rates decreased as the food concentration increased over \( 4 \times 10^5 \) cells (or \( 1.5 \times 10^{-2} \) mg C)/ml. Below this concentration, the rates in the experiments of 2-3 hour duration were in many cases higher than
The food removal rates by *N. scintillans* in various levels of suspensions of *T. tetrathelle* and *G. nagasakiense*. The open circles represent the experiments of 2-3 hour duration and the solid circles the experiments of 4-7 hour duration.

Fig. 2.

The food removal rates by *N. scintillans* in various levels of suspensions of *T. tetrathelle* and *G. nagasakiense*. The open circles represent the experiments of 2-3 hour duration and the solid circles the experiments of 4-7 hour duration.

Fig. 3.

The food removal rates by *N. scintillans* in various levels of suspensions of *G. nagasakiense*. The open circles represent the experiments of 2-3 hour duration and the solid circles the experiments of 4-7 hour duration.

Fig. 4.

The food removal rates by *N. scintillans* in various levels of suspensions of *G. nagasakiense*. The open circles represent the experiments of 2-3 hour duration and the solid circles the experiments of 4-7 hour duration.

The food removal rates were calculated using the mean food concentrations, but plotted in Figs. 3 and 4 against the initial food concentrations as suggested by McClatchie and Lewis. The food removal rates were calculated using the mean food concentrations, but plotted in Figs. 3 and 4 against the initial food concentrations as suggested by McClatchie and Lewis. The food removal rates were calculated using the mean food concentrations, but plotted in Figs. 3 and 4 against the initial food concentrations as suggested by McClatchie and Lewis.
rate increased with increasing food levels. The rates in the experiment of 2-3 hour duration were generally higher than those of 4-7 hour duration, but the maximal rates were nearly the same, except one case, at about 2000 cells (or \(0.75 \times 10^{-4} \text{mg C}/\text{Noctiluca/h}\). This maximal rate was reached in both cases around \(4 \times 10^5\) cells/ml, and there were no further increases with increasing food levels. Fig. 4 shows the food removal rates with \(G. \text{nagasakiense}\). The rates also increased with increasing food levels, and the maximal rate of 600 cells (or \(1.5 \times 10^{-4} \text{mg C}/\text{Noctiluca/h}\) was reached around \(1 \times 10^5\) cells/ml.

**Discussion**

The results of the experiments with \(T. \text{tetrathelle}\) (Fig. 2) demonstrate that the clearance rate is the function of both food concentration and experimental duration. The feeding of \(\text{Noctiluca}\) in this experiment seems to have occurred soon after the start of experiment to see that the algal masses appeared in the body of \(\text{Noctiluca}\) twenty minutes after the inoculation. The food concentration of \(4 \times 10^5\) cells/ml, where the clearance rates started to decline, is generally consistent with the concentration where the maximal growth rate of \(\text{Noctiluca}\) was obtained. The result that the higher clearance rates were obtained in the experiments of shorter duration than in the longer presents two possible causes. One is that the starved \(\text{Noctiluca}\) secreted mucus more actively in the earlier period of exposure to food suspension, and the other is that they fed more actively in that period. The result in Fig. 3 that the maximal food removal rates were nearly the same regardless of the experimental duration implies that the maximal rate obtained here is generally equal to the limit of food removal ability in \(\text{Noctiluca}\). Though the clearance rates with \(T. \text{tetrathelle}\) and \(G. \text{nagasakiense}\), when compared on the basis of experimental duration, showed no clear differences between them, the maximal food removal rates (Figs. 3 and 4) were quite different from each other in terms of both cell number and amounts of carbon. This is supposed to be due to the difference in cell size: The size of \(T. \text{tetrathelle}\) was \((10-20) \times 20 \mu\text{m}\) and that of \(G. \text{nagasakiense}\) was \((25-30) \times (28-32) \mu\text{m}\).

When the maximal food removal rates were expressed in terms of cell volumes, which were roughly calculated by cell length \(\times\) width \(\times\) thickness, they were \(4 \times 10^{-3} \text{mm}^3/\text{Noctiluca/h}\) with \(T. \text{tetrathelle}\) and \(7 \times 10^{-3} \text{mm}^3/\text{Noctiluca/h}\) with \(G. \text{nagasakiense}\): The ratio of cell thickness to cell width is about 2:3 for \(T. \text{tetrathelle}\) and about 1:2 for \(G. \text{nagasakiense}\). Assuming that \(\text{Noctiluca}\) is a ball with a radius of 400 \(\mu\text{m}\), its volume is about \(3 \times 10^{-2} \text{mm}^3\), and the maximal volume of food it removes an hour is roughly equivalent to \((1/8-1/4)\) of its volume.

The maximal concentration of \(G. \text{nagasakiense}\) in the sea reached \(3 \times 10^9\) cells/liter, which is far higher than the concentrations examined in this study. This raises a possibility that \(\text{Noctiluca}\) may inflict feeding pressure on the growth of \(G. \text{nagasakiense}\) in the sea, resulting in the suppression of red tide it induces. The number of \(\text{Noctiluca}\) needed to suppress the population growth of \(G. \text{nagasakiense}\) can be roughly calculated using the following equation:

\[
dC/dt = k \cdot C - F \cdot C \cdot N,
\]

where \(C\) is the concentration of \(G. \text{nagasakiense}\) (cells/ml), \(t\) is time (day), \(k\) is the specific growth rate of \(G. \text{nagasakiense}\), \(F\) is the clearance rate by \(\text{Noctiluca}\) (ml/\text{Noctiluca/day}), \(N\) is the density of \(\text{Noctiluca}\) (inds./ml). The diel clearance rate by \(\text{Noctiluca}\) will be lower than simple 24 fold of the hourly rate obtained in this study, because \(\text{Noctiluca}\) stops feeding activity during the cell division. As it takes 6-8 hours to complete the cell division and the division rate also varies depending on the environmental conditions, the diel rate in this calculation was assumed to be about ten fold of the hourly rate.
Taking as an example $F = 4 \times 10^{-2}$ ml/Noctiluca/day, $k = 0.7$ (G. nagasakiense divides nearly one time a day in the laboratory culture)$^{16}$, $C = 5 \times 10^4$ cells /ml, 18 Noctiluca/ml will be needed to stop the population growth of G. nagasakiense. But, as this study was conducted under constant environments in the laboratory, confirmative field studies are necessary to apply the results obtained here to the phenomena occurring in nature.

**References**


* : in Japanese with English summary
** : in Japanese
夜光虫（Noctiluca scintillans）の Tetracelmis tetrathelle と Gymnodinium nagasakiens 懸濁液中での餌料除去率

李 正根・平山 和次

夜光虫は触手の周りに粘液を分泌して餌料を集め摂取する。懸濁液中で摂取する生物に用いられる餌料密度の減少がそれに影響する摂取量を求める方法を応用して、いろいろな餌料密度を変化させた時間で夜光虫を飼育し、その餌料密度の減少から、みかけの懸濁液速度、餌料除去速度を求めた。T. tetrathelle を飼料とすると、餌料密度が 4 × 10^6 cells/ml 以下の時、短い実験時間（2-3 時間）で求めた最高懸濁速度（約 2 × 10^{-2} ml/Noctiluca/h）は長い実験時間（4-7 時間）で求めたものには比べて 2 倍であった。餌料密度が 4 × 10^6 cells/ml 以上では懸濁速度は低下した。また G. nagasakiens を餌料とした時（実験時間は 4-5 時間）、懸濁速度は 1 × 10^{-3} と 7 × 10^{-3} ml/Noctiluca/h の間で変動した。どちらの餌料の場合でも餌料除去速度は餌料密度の増加とともに増加し最大値に達した。それ以上の餌料密度では餌料除去速度の増大は認められなかった。最大餌料除去速度は T. tetrathelle の場合は 2000 cells （または 0.75 × 10^{-3} mg Carbon)/Noctiluca/h であり、G. nagasakiens の場合は 600 cells （または 1.5 × 10^{-4} mg Carbon)/Noctiluca/h であった。海域で夜光虫の密度が高い場合には、その増殖が G. nagasakiens の増殖を抑制する可能性は充分考えられる。