The Effect of Light Intensity and Temperature on the Growth of Several Marine Algae useful for rearing Molluscan Larvae

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The Effect of Light Intensity and Temperature on the Growth of Several Marine Algae useful for rearing Molluscan Larvae

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Summary

1. The effect of light intensity and temperature on the growth of four algae used for rearing molluscan larvae was investigated.

2. Growth was measured by direct count of the increase in cell concentration. The normal growth constant was used as an index in comparing the growth rate under different environmental conditions.

3. The optimum conditions of light intensity and temperature for the growth of four algae were as follows.

Monochrysis lutheri	4,500– 8,000 lux,	20–25°C.
Platymonas sp.	4,500– 8,000 lux,	23–25°C.
Nitzschia closterium	4,500–12,500 lux,	23–29°C.
Chaetoceros calcitrans	4,500–12,500 lux,	23°C.

4. *Chaetoceros calcitrans* was found to grow very much faster than the three other algae under optimum conditions, suggesting the advantage of using the species for the rearing of molluscan larvae on a large scale.

Recently, a technic for rearing the larvae of various marine animals for the purpose of obtaining their seeds has been developed (2, 6, 7). At the same time, marine algae, such as flagellates and diatoms, have been cultured on a large scale to provide food for the molluscan larvae and juveniles (1, 3, 14).

Regarding the culture of algae, the relation between the growth of algae and the light intensity and temperature has been studied by many scientists. Tamiya et al. (11) analyzed the growth of Chlorella at various light intensities and temperatures. Sorokin and Krauss (8, 9) also studied the effects of light intensity and temperature on the growth of Chlorella. Talling (10) studied the growth of diatoms under natural and laboratory conditions. Ukeles (12) studied the effect of temperature on the growth of several marine algae and Jitts et al. (4) studied the division rates of some marine phytoplankters at various light intensities and temperatures.

In the culture of larvae and juveniles of mollusca on a large scale, it is of prime importance to increase the efficiency of the algal production by keeping the culture under optimum conditions. The present research was planned to find the optimum condition of temperature and light intensity for the growth of several algae which had been widely used for mass culture of molluscan larvae and juveniles.

Experimental methods

Species of algae used in the study were Chrysophyta; *Monochrysis lutheri*, Chlorophyta; *Platymonas sp.*, Bacillariophyta; *Nitzschia closterium* and *Chaetoceros calcitrans*. *Monochrysis* has been widely used as food organism for the rearing of molluscan larvae (1, 14). *Platymonas sp.* is an effective food organism for the early juvenile of abalone (3). *Nitzschia* has been a popular alga used in the culture of many marine animal larvae. *Chaetoceros calcitrans* was isolated in Japan (13) and since then has been widely used as food for many molluscan larvae (5). Imai (3) made a successful rearing of several molluscan larvae by using a mixture of *Monochrysis* and *Chaetoceros calcitrans*.

The culture was grown in 25 ml test tubes which were kept in a water bath at a constant temperature. The bath container was made of transparent vinyl chloride plate (1 mm thick). Twenty W. day-light fluorescent lamps (Mitsubishi Electric Company) were used as light sources and illuminated the sides of the water bath. The light intensities were measured by an illuminometer (Toshiba Electric Company, Type No. 5).

TABLE 1. *Composition of Culture Medium Used.*

Dist. water	1000 ml
NaCl	18 g
KCl	600 mg
$NaNO_3$	500 mg
$MgSO_4 \cdot 7H_2O$	5 g
Ca (as Cl^-)	100 mg
K_2HPO_4	30 mg
* PI metals	30 ml
Fe (as Cl^-)	100 γ
Tris	1 g
Vit. B_{12}	3 γ
Na_2SiO_3	80 mg
** Vit. I	1 ml
pH	7.5

* 100 ml=Na_2EDTA 100 mg, Fe 1 mg, Zn 0.5 mg, Mn 4mg, Co 0.01 mg, Cu 0.004 mg, B 20 mg.
** 50ml=Vit. B_{12} 10γ, Biotin 50γ, Vit. B_1 5 mg.

The composition of the synthetic culture medium is shown in Table 1. The medium was autoclaved at 1 kg/cm^2, for 20 minutes. Cultures for the experiment were prepared in 100 ml Erlenmeyer flasks containing about 60 ml of sterilized medium and grown for about 70 hours at 20±1°C under continuous illumination

(6,500 lux). Cultures, thus prepared, were in the exponential phase of growth. The cells, which were separated by a centrifuge at 1,600–1,700 rpm for 20 minutes, were inoculated into the test tubes for culture experiment. A sample of one tube was taken at intervals and the cell number was counted with a hemacytometer.

In the case of *Platymonas* and *Nitzschia* the cells attach themselves to the substratum. Therefore, the culture was vigorously stirred several times with a sterilized pipette before sampling. Five counts were made for each culture. When the cell number was plotted on semilogarithmic paper, a straight line was obtained between the cell count and the time. The growth constant (k) was calculated from the slope of the straight line by an equation $k=\log N_1-\log N_0/t_1-t_0$ as is shown in Fig. 1. The constant (k) was used as a norm in comparing the rates of growth under different environmental conditions.

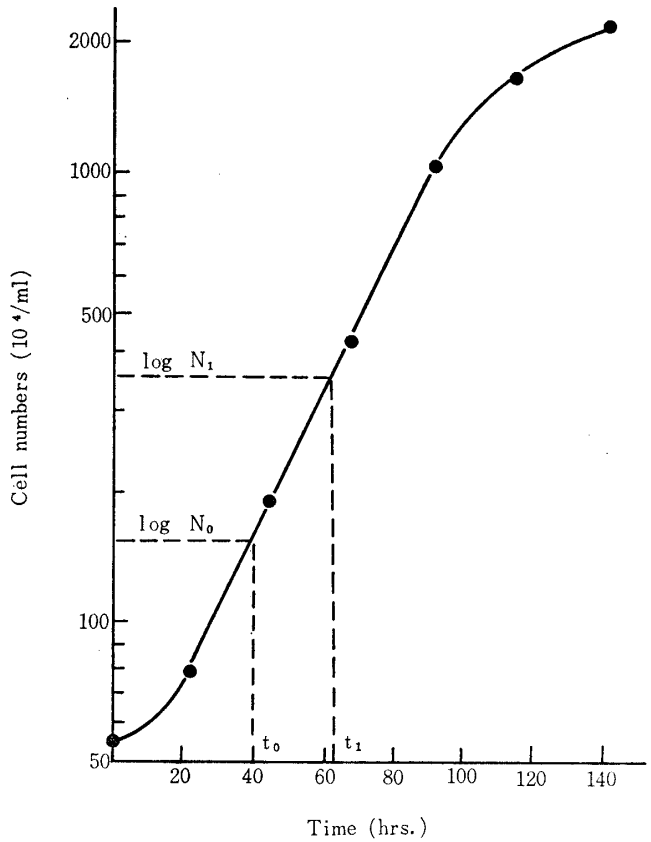

FIG. 1. Growth curve of *Monchrysis* at continuous illumination (4,500 lux), 20±1°C.

Results

1. *Monochrysis lutheri*

The growth constant (Fig. 2) increased proportionally to the intensity of light up to about 4,500 lux at each temperature and then slowly declined as the intensity of light increased. The decline at the higher light intensity was rather

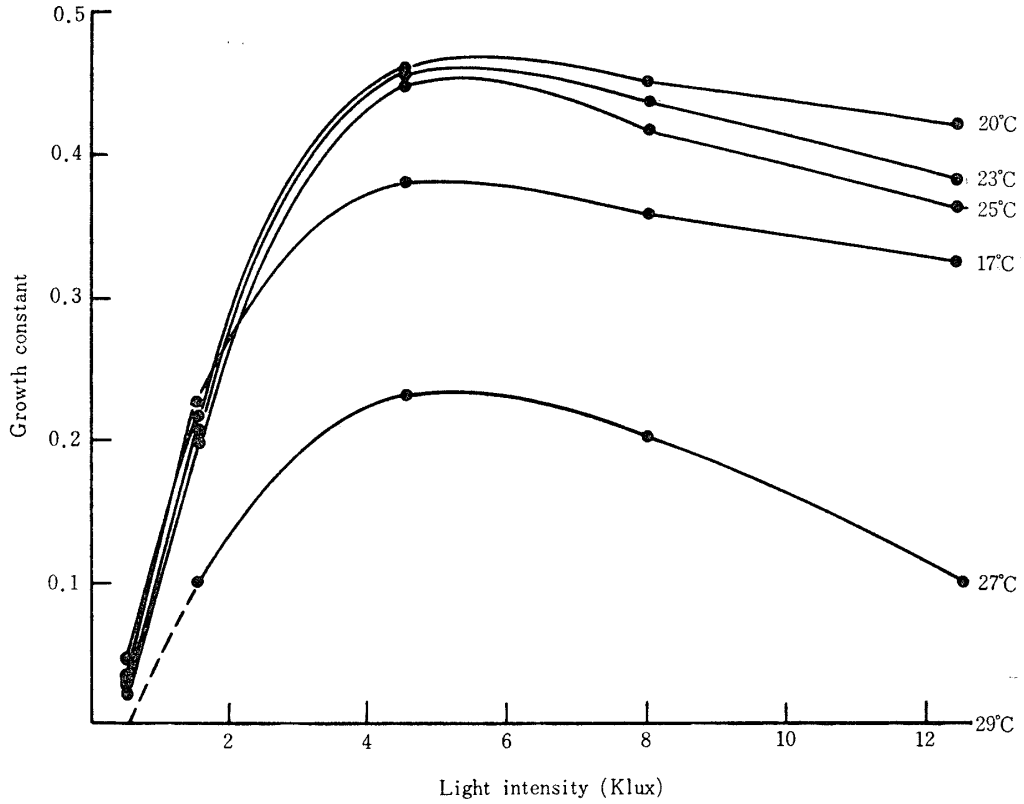

FIG. 2. Growth constant curves of *Monochrysis lutheri* cultured at various light intensities and temperatures.

remarkable at 27°C. There was little variation in the growth constant between 20° and 25°C. At 27°C, the growth constant was distinctly lower and the alga disappeared at the lowest illumination. At 29°C, the growth constant was zero at every temperature. The alga could not survive at this high temperature. Therefore, it is clear that the upper limit of temperature for the growth of the species is between 27° and 29°C.

2. *Platymonas sp.*

The cruve of the growth constant was different in shape at different temperature conditions. That is, at 23–29°C, the constant increased in accord with the light intensity up to 4,500 lux and then a gradual decline as the light increased up to 12,500 lux. While at 17–20°C, the constant increased continuously up to the light intensity of 12,500 lux. Though the growth constant was not measured at light intensities over 12,500 lux at this lower temperature condition, it seems reasonable from Fig. 3, that for growth of the species, the optimum conditions are 4,500–8,000 lux and 23–25°C.

3. *Nitzschia closterium*

The growth constant curves (Fig. 4) were alike at every temperature, although

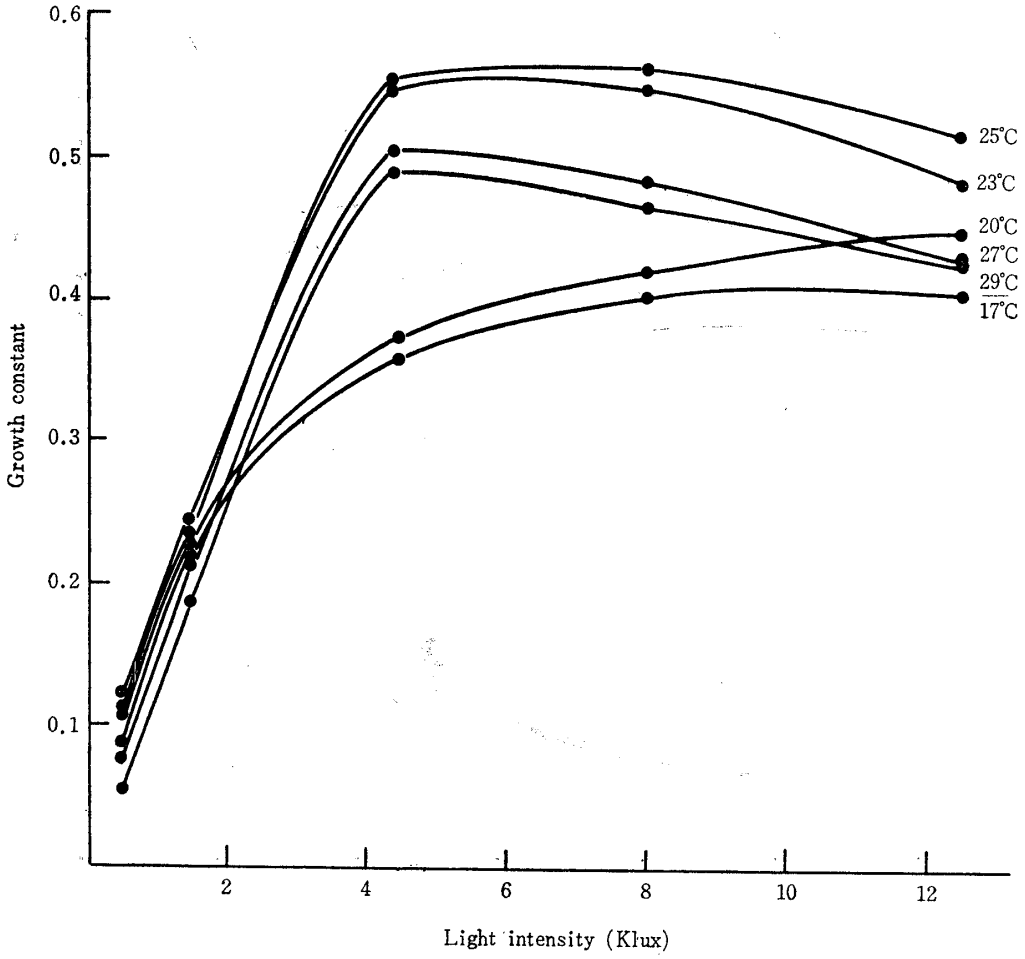

FIG. 3. Growth constant curves of *Platymonas sp.* cultured at various light intensities and temperatures.

the value of the constant was different at each temperature. The constant increased up to 4,500 lux and then declined slowly as light intensities increased. Between 23–29°C, the constants were the highest with a small variation. At a lower temperature, the constant was significantly low. Therefore, the optimum conditions for growth of this species is estimated to be 4,500–12,500 lux and 23–29°C.

4. *Chaetoceros calcitrans*

The forms of the growth constant curves were alike at every temperature condition (Fig. 5). The constants increased proportional to the increase of light intensity up to 4,500 lux and then, stayed at the same level up to 12,500 lux. As to the temperature, the constants increased rapidly as the temperature increased up to 23°C and then decreased rapidly. The constant at 23°C was twice as high as that at 17°C. The optimum temperature condition for the growth of the species is around 23°C and the optimum light intensity is 4,500–12,500 lux.

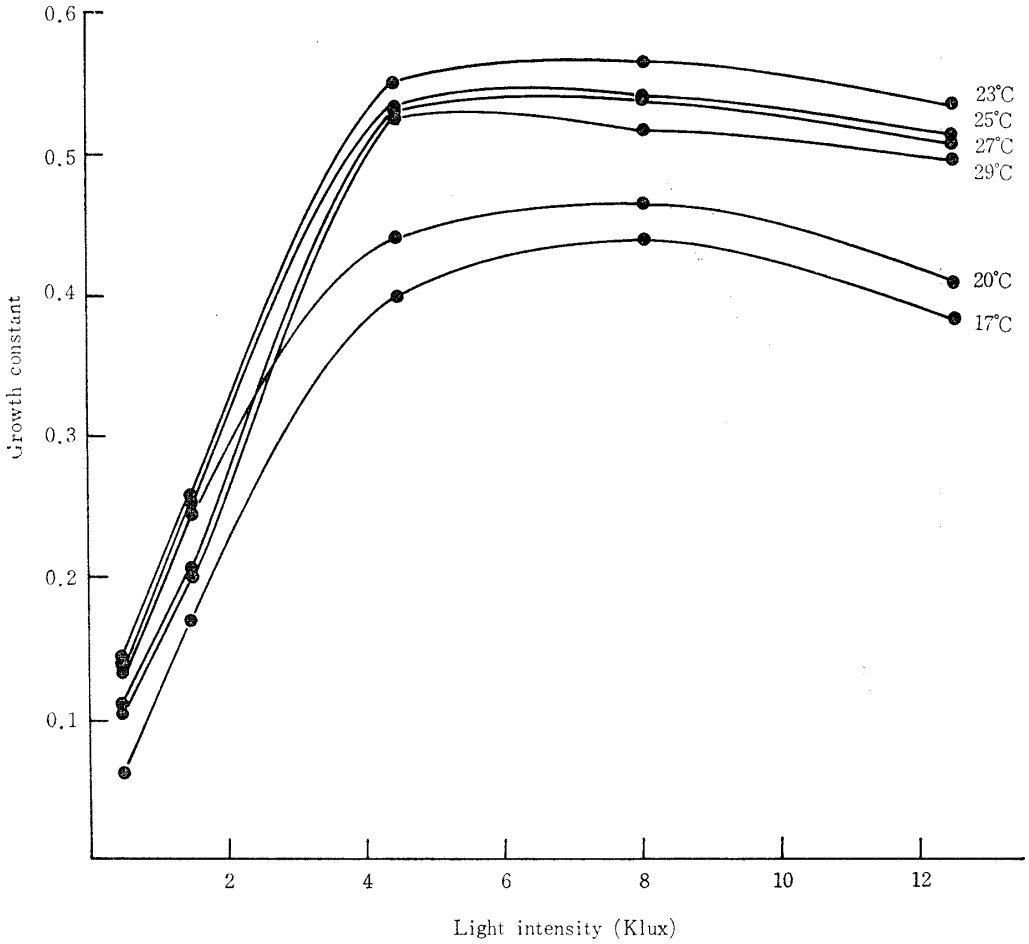

FIG. 4. Growth constant curves of *Nitzschia closterium* cultured at various light intensities and temperatures.

Discussion

Among the algae used in this experiment, *Monochrysis* had been the one studied rather extensively regarding the light and temperature relation in growth. Jitts et al. (4) found the optimum temperature for the growth of *Monochrysis*, at 19°C, a little lower than the present result. Ukeles (12) found, in *Monochrysis*, a distinct inhibition at 27°C and no growth at 29°C. Our results are in good agreement with hers (Fig. 2). The present data on the three other algae seem to be new to science.

The characteristics of growth in relation to light intensity were similar among the four algae studied. Namely, the highest values of the growth constants were obtained under the illumination between 4,500 and 8,000 lux. The rate increased rapidly up to the intensity of 4,500 lux and then stayed at almost the same level up to 8,000 lux. Therefore, for the sake of producing these algae most efficiently, the illumination of around 4,5000 lux would be preferable.

The growth constants under optimum environmental conditions in *Mono-*

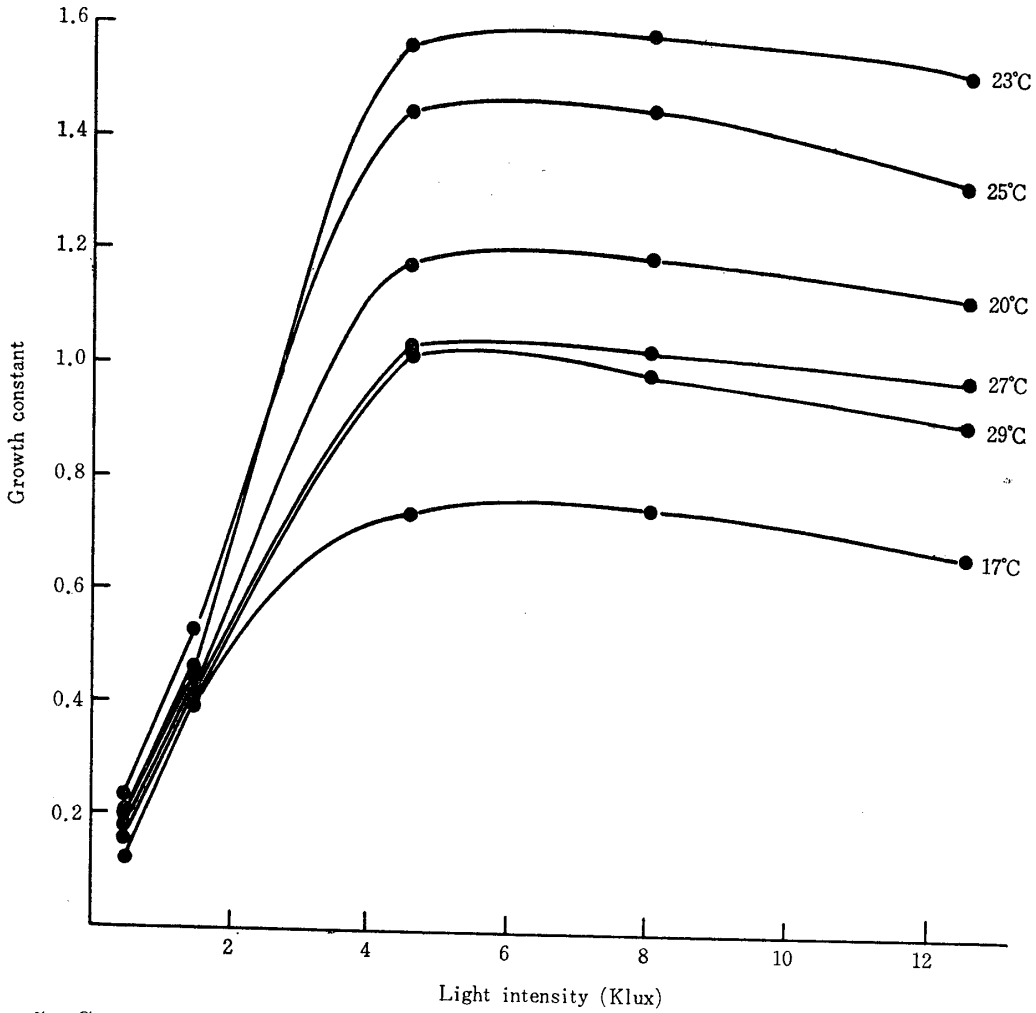

FIG. 5. Growth constant curves of *Chaetoceros calcitrans* cultured at various light intensities and temperatures.

chrysis lutheri, *Platymonas sp.* and *Nitzschia closterium* were of the same order between 0.45 and 0.55 while it was 1.50 in *Chaetoceros clacitrans*. That means it grows very much faster than the other three. Therefore, *Chaetoceros calcitrans* is considered as the best of the four algae studied regarding the rate of growth.

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