Utilization of Nitrate & Phosphate by the Green Alga
Tetraselmis gracilis Kylin

S. Z. QASIM

National Institute of Oceanography, Dona Paula 403004

&

K. J. JOSEPH

Central Marine Fisheries Research Institute, Cochin 682018

Nutrient-depleted cells of *T. gracilis* were grown in sea water containing different concentrations of nitrate and phosphate. Growth rate of cells was first studied by determining the growth constant (k) and generation time (t_g). Enrichment with nitrate and phosphate either singly or in combination increased k and reduced t_g values. Further studies on algal growth were made using the Monod model. Cell counts and ^{14}C uptake were taken as indices of growth parameter (μ). Values of half saturation constant (K_s) and maximum growth rate (μ_{max}) obtained from cell counts varied from those of ^{14}C uptake. Certain modifications in the Monod model seem necessary for the study of algal ecology and these modifications should include multiple variability of the environment and the specific behaviour of the algae.

RECENT studies on the rate of uptake of nitrate and phosphate by the nutrient-depleted cells of a diatom, *Biddulphia sinensis*, and a dinoflagellate, *Ceratium furca*, provided some evidence of their growth kinetics[1]. Similar investigations were carried out on a green alga, *Tetraselmis gracilis*, in which growth constant and generation time were determined in relation to nutrient concentrations. Effects of nutrients on algal growth were demonstrated using the Monod model which states

$$\mu = \mu_{max}\left(\frac{S}{K_s+S}\right)$$

where μ is the specific growth of the algae, μ_{max} is the maximum growth rate unlimited by low nutrient concentration, S is the nutrient concentration and K_s is the half saturation constant which is equal to $\mu_{max}/2$.

Materials and Methods

Unialgal cultures of *T. gracilis* were grown in the laboratory until they became nutrient-limited. Sea water samples of known concentrations of salinity, nitrate and phosphate were then taken in a series of conical flasks (5 l). The sea water used was filtered and made sterile. Each flask was then enriched with nitrate-nitrogen so that the 1st set of 5 flasks had concentrations of 0·5, 1, 2, 5 and 10 µg-at/litre respectively in addition to the initial concentration. Similarly, the 2nd set of 7 flasks was enriched with phosphate-phosphorus having 0·5, 1, 2, 3, 5, 6 and 10 µg-at/litre. In the 3rd set of 5 flasks, both phosphate and nitrate were used in combination. The concentrations of these two nutrients combined were 1, 2, 5, 10 and 20 µg-at/litre. One flask with each

set was kept as unenriched and used as control. Equal volumes of *T. gracilis* were then added in each set of flasks and the cultures exposed to 8 hr of fluorescent illumination (20 klux) during the day, followed by darkness. A gentle stream of air bubbles was passed through all the flasks continuously to keep the organisms in suspension.

Small quantities of cellular material were pipetted out from each flask daily for the cell counts. Aliquots of 50 ml were drawn from each flask on every alternate day and their rates of photosynthesis were measured by ^{14}C uptake.

Growth constant and generation time were calculated from the well known equations

$$\ln.n_t = \ln.n_0 + kt$$
$$\text{or} \quad \ln p_t = \ln.p_0 + kt$$
$$t_g = 0·7/k$$
$$\text{or} \quad k = 0·7/t_g$$

where n_t or p_t and n_0 or p_0 are the cell counts at times t and 0 respectively and k is the growth constant expressed in hr^{-1} and t_g is the mean generation time expressed in hours.

Results and Discussion

Growth Constant and Generation Time

One of the simple approaches to study the growth of unialgal cultures is by determining the growth constant (k) and generation time (t_g). The experimental populations show an initial lag phase in growth followed by vigorous logarithmic growth phase and finally a decline in growth introduced as a result of depletion of nutrients or trace elements. Growth constant (k) is a measure of total metabolism of the algae and the mean generation time (t_g) is the average time taken for the cells to divide. The former lies within the range of