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The impact of the Cyanobacterium *Synechocystis salina* on *Chaetoceros affinis*

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Abstract

The characteristic growth pattern, chlorophyll concentration and productivity of a cyanobacterium *Synechocystis salina* and a diatom *Chaetoceros affinis* have been studied separately. The effect of *S. salina* on the growth, pigments and primary productivity of *C. affinis* has been assessed. An inhibitory effect has been noticed in the growth, concentration of chlorophyll *a* and *c* , and in the net production while an enhancing effect has been observed in the phaeophytin concentration of the diatom. The mutual impact of the species of two different classes has been assessed in their dispecific culture under the same physicochemical parameters. An overall inhibition on the growth, chlorophyll concentration and productivity was observed.

Certain algae, abundant in fresh and sea water have attracted attention because of their lethal effects on fish and other animals. Ballantine and Abbott (1957) reviewed the occurrence and physiological effects on animals of toxic marine flagellates- *Gymnodinium* and *Gonyaulax* are most prominent. The toxin of *Gymnodinium veneficum* can kill various molluscs, fish and mammals by acting specifically on the nervous system. The brackish-water chrysophyte *Prymnesium parvum,* which is occasionally responsible for mass mortality of fish, produces a nondializable, acid-labile and thermolabile extracellular toxin (Shilo and Aschner, 1953; McLaughlin, 1958). Domestic birds and mammals die occasionally as a consequence of drinking water containing dense blooms of cyanobacteria, among which species of *Microcystis* and *Anabaena* seem to be particularly poisonous (Ingram and Prescott, 1954; Gorham, 1960). Four toxic factors from algal blooms have now been recognized on the basis of the speed at which they act and the symptoms they produce toxins: hepatotoxic peptides, neurotoxic alkaloids and dermatotoxic phenolic compounds (Carmichael, 1988; Codd and Poon, 1988) in addition to lipopolysaccharide (LPS) endotoxins (Drews and Weckesser, 1982; Keleti and Sykora, 1982). Hepatotoxic and neurotoxic blooms have caused animal poisonings all over the world (Skulberg et al., 1984; Gorham and Carmichael, 1988).

It may be suggested that toxic cyanobacteria may also be a health hazard for humans (Bourke and Hawes, 1983; Carmichael et al., 1985; Codd and Poon, 1988). The effect of cyanobacteria on members of plant kingdom has also been studied. A secondary metabolite (cyanobacterin) produced by the cyanobacterium *Scytonema hofmani* was

shown to inhibit the growth of algae. It also inhibited the growth of angiosperms including *Lemna,* and terrestrial species such as corns and peas (Florence and Deborah, 1985). It might be used as a commercial algicide for algal waterbloom control. Inspite of such intensive studies, the impact of cyanobacteria on microalgae has not been investigated so far. Investigations show that extracellular products inhibit their own growth or that of other species. Autoinhibition of growth may be due to the accumulation of metabolic products in the medium as was observed in the case of *Nostoc punctiforme* (Harder, 1917). Jorgensen (1956) analysed the growth inhibiting substances produced by a few algal species. Several workers have reported that certain species of algae inhibit the growth of other algae in synspecific cultures. Flint and Moreland (1946) observed antibiosis in a few species of cyanobacteria.

Proctor (1957) demonstrated algal antibiosis using *Haematococcus* and *Chlamydomonas* species. Rice (1954) studied the impact of bioactive extracellular product on the population of planktonic algae. The aim of the work is to study the impact of the cyanobacterium *Syreclocyster salina* on the growth, pigment concentrations and primary productivity of *Choetoceros affinis.*

Material and methods

A known volume of culture medium was taken in each of the three conical flasks of 2 litre capacity. To the first flask a known concentration of *Synechosystis salina* culture was inoculated and to the second one a known concentration of *Chaetoceros affinis* culture was added. The third flask contained a known volume of definite concentration of *S. salina* and *C. affinis* were added. All the three flasks were exposed to intermittent light (fluoresecent tube) and dark periods of 12 hrs each. Cell concentration, oxygen evolution and pigment concentration in all the three flasks were estimated at regular intervals.

S.salina filtrates of known volume was collected on the seventh and fourteenth days and were added to fresh *C.affinis* cultures, the control of test organism being maintained. The same physicochemical parameters were observed.

Cell concentration was measured using a Haemocytometer. Pigments were measured spectroscopically using standard methods (Strickland and Parsons, 1972). Primary productivity measurements were made using Gaarder and Gran's light and dark bottle method as described by Strickland and Parsons (1972) and APHA (1992)

Results and discussion

The generation time of *C.affinis* alone and when cultured along with *S. salina* is shown in Table 1. The growth was found inhibited in dispecific culture and also when grown with the filtrate from fourteen days old *S.salina* culture. In the unialgal culture of *C.affinis* after 24 hrs. of incubation the generation time (tg) was 25 hrs. When the cyanobacteria was inoculated the growth of the diatom was arrested for the first 48 hrs.. The dispecific culture it took 72 hrs. to initiate any cell

Table 1. *Effect of S. salina culture on generation time of C. affinis.*

No of days	Control (tg in hours) *C. affinis* alone culture	With *S. salina* (tg in hours)
1	25.0	-
2	31.8	-
3	37.3	233
4	37.1	175
5	-	-
6	43.3	-
7	47.8	140
8	43.5	205
9	49.9	350
10	57.5	350
11	61.2	233
12	65.5	233
13	65.6	700
14	70.2	700

multiplication when the tg value recorded was 23 hrs. Corresponding tg for the monospecific culture was only 37 hrs. on the third day. In the dispecific culture on the fourteenth day the tg was 700 hrs. as against 70 hrs. in the monospecific culture This conspicuous delay in the cell multiplication is due to the cyanobacterial inhibiting impact on the diatom. This negative impact was projected even at a later stage of growth. In monospecific culture though the growth continued at a lower rate recording 133 hrs. On the 25th day in dispecific culture the growth was totally arrested for the 20th day onwards rendering the cell nonviable.

The monospecific culture of *C. affinis* exhibited gradual increase of chlorophyll *a* from 43.8 µg/l to 145.6 µg/l on the eleventh day. Filtrate from seven days old culture and 14 days old culture of cyanobacteria also showed increase in chlorophyll but of lower magnitude (Table 2. a & b). On the eleventh day corresponding chlorophyll *a* values for seven days and fourteen days old filtrate being 130.4µg/l and 68.15 µg/l. The chlroophyll a:c ratio in monospecific culture of *C. affinis* was 8:5. *C. affinis* with 7 days old filtrate of *S. salina* allowed this ratio to 1:2 and 14 days old filtrate for the same *a* : *c* ratio of 2:1.

The average chlorophyll *a* : phaeopigment value was found to be 1:7 in the monospecific culture; with seven days old filtrate of *S. salina* the ratio was found to be 1:8 indicating the increase in the concentration of phaeopigments and decrease in chlorophyll *a* concentration. The filtrate from the fourteen days old culture further altered the chlorophyll *a* : phaeopigment ratio to 1:2.

The chlorophyll *a* : carotenoid ratio was also found altered under the influence of the cyanobacterium while the monospecific culture registered chlorophyll *a* : carotenoids ratio as 3:2. *C. affinis* with 7 days old and 14 days old filtrate from cynobacterium gave the altered ratio of 1:1 and 1:3 respectively (Table 2 c).

The productivity potential of the diatom was very much affected by the cyanobacterial impact as evidenced by the considerable decrease in the chlorophyll *a* concentration (Table 2. d and e). The productivity of the diatom *C. affinis* was also found to be inhibited by the cyanobacterium *S. salina* probably representing the productivity potential cut in the nature. When the average productivity of

Table 2.

a Effect of seven days old S. salina filtrate on chlorophyll a concentration of C. affinis.

No. of days	Control (µg/litre)	Wtih 7 days old filtrate of *S. salina* (µg/litre)
0	1.13	1.51
4	26.40	29.2
8	81.93	50.73
13	86.46	68.15
19	324.91	227.57

b. Effect of fourteen days old S. salina filtrate of chlorophyll c concentration of C. affinis

No. of days	Control (µg/litre)	Wtih 7 days old filtrate of *S. salina* (µg/litre)
0	10.54	10.61
4	22.06	23.52
8	74.52	33.18
13	65.53	55.22
19	192.03	147.79

c. Effect of fourteen days old S. salina filtrate on carotenoids concentration of C.affinis.

No. of days	Control (µg/litre)	Wtih 7 days old filtrate of *S. salina* (µg/litre)
0	2	1.6
4	14	18
8	56	40
13	71.2	70.4
19	187.6	114.8

d. *Effect of seven days old S. salina filtrate on productivity of C. affinis.*

No. of days	Control (µg/litre)	Wtih 7 days old filtrate of *S. salina* (µg/litre)
0	0.911	0.871
4	0.458	1.251
8	1.452	2.367

e. *Effect of fourteen days old S. salina filtrate on productivity of C. affinis.*

No. of days	Control (µg/litre)	Wtih 7 days old filtrate of *S. salina* (µg/litre)
0	0.625	0.179
4	0.581	0.313
8	0.983	0.625

the monospecific culture without the filtrate of the seven days old *S. salina* culture gave the average value of 1.17 mgC/l/hr., the diatom with the filtrate recorded the average production of 0.45 mgC/l/hr. This one-third cut in productivity and productivity potential is a grave problem in nature which adversely affects the final resources.

The observations indicate that the cyanobacterium *S. salina* produces bioactive substances which inhibit the growth of the diatom *C. affinis*. This inhibiting effect was quite obvious as is expressed in the decrease in the cell numbers, chlorophyll concentration and photosynthetic rate. Growth inhibition among several plankton species by *Scytonema hofmani*, a freshwater cyanobacterium was reported by Mason *et al.* (1982). Either the intact filaments or cell extracts of *Scytonema hofmani* was found to have inhibited several bluegreen algal species such as *Synechococcus sp., Anacystis nidulans, Microcystis aeruginosa, Anabaena cylindrica and Nostoc commune,* and green algae such as *Cosmarium botrytis, Chlamydomonas reinhardii* and *Chlorella pyrenoidosa.* In the present investigations the inhibitory effect of metabolites from *Synechocystis salina* was found to have considerable inhibitory effect so as to annihilate the coexisting species.

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