

2096  
(86)



PRIMARY PRODUCTION AND PHOTOSYNTHETIC ACTIVITY OF 3  
PHYTOPLANKTON SPECIES CULTIVATED UNDER HIGH ZN  
CONCENTRATIONS

Luciana Andrade<sup>1,2</sup>; Sandra M.F.O. Azevedo<sup>2</sup> and  
Wolfgang C. Pfeiffer<sup>1</sup>

<sup>1</sup>Instituto de Biofísica Carlos Chagas Filho

<sup>2</sup>Núcleo de Pesquisas em Produtos Naturais

Universidade Federal do Rio de Janeiro

CCS - Cidade Universitária - Ilha do Fundão - 21949-900  
Rio de Janeiro -RJ - Brasil

**SUMMARY:** Three phytoplankton species were cultivated in different Zn concentrations (0.08 - 8.00  $\mu\text{M}$ ). Primary production and photosynthetic activity were determined by  $\text{NaH}^{14}\text{CO}_3$  incorporation. Photosynthetic activity increased up to 74% under 4.00 and 8.00 $\mu\text{M}$  of Zn. This can be understood as algae self-protection in order to overcome pollution.

**KEY WORDS:** Primary production, Photosynthetic Activity, Phytoplankton, Zn.

INTRODUCTION

Many aquatic environments in different areas of the world are polluted with high heavy metals concentrations<sup>1</sup>. In Brazil heavy metal contamination has been well documented for Sepetiba Bay, State of Rio de Janeiro, due to a Zn and Cd industry<sup>2,3,4,5</sup>.

In heavy metal polluted waters, phytoplankton has to develop physiological adaptations to survive. Among these changes some are very specific like the production of metallothioneins<sup>6</sup> or special excretion mechanisms<sup>7</sup>. Probably, before the occurrence of such modifications, some others can occur. In algae they may involve adaptations in photosynthesis process in order to maintain the energy supply.

In the present paper three phytoplankton species from Sepetiba Bay were cultivated with different Zn

concentrations to observe any possible change in primary production and photosynthetic activity.

#### MATERIALS AND METHODS

*Phaeodactylum tricornutum* (diatom), *Chlorella* sp. (chlorophyte) and *Synechocystis* sp. (cyanobacteria) were cultivated respectively in f/2, WC and ASM-1 media<sup>8,9,10</sup>. Simultaneous cultures with 0.08  $\mu\text{M}$  (control); 0.16  $\mu\text{M}$  (20 Zn); 4.00  $\mu\text{M}$  (50 Zn) and 8.00  $\mu\text{M}$  (100 Zn) were maintained under  $25 \pm 2^\circ\text{C}$  and  $67 \mu\text{E} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$  in a 14:10 light: dark cycle. In exponential phase primary production was measured by  $\text{NaH}^{14}\text{CO}_2$  incorporation<sup>11</sup> using a total activity of  $74 \times 10^4 \text{Bq}$  (20  $\mu\text{C}$ ). After a 4 hour period of incubation, cultures were filtered in 0.45  $\mu\text{m}$  Millipore cellulose membrane. Filters were dissolved in Bray solution<sup>12</sup> and samples were counted in a LS-250 Beckman Scintillation System. The same cultures were monitored for chlorophyll *a* measurements following extraction in acetone 90% and spectrophotometric readings at 665 nm and at 750 nm for turbidity correction<sup>13</sup>.  $^{12}\text{C}$  correction by alkalinity and results conversion to  $\mu\text{g C} \cdot \text{l}^{-1} \cdot \text{h}^{-1}$  or to  $\mu\text{g C} \cdot \text{chl} \cdot \text{l}^{-1} \cdot \text{h}^{-1}$  also followed<sup>13</sup>. Final results were compared by means of percentage related to the control (100%).

#### RESULTS AND DISCUSSION

Table 1 shows primary production values for the 3 species studied. Comparing results for  $10^6$  cells, no significant difference was found for the diatom cultivated under high Zn concentrations. However for the chlorophyte and the cyanobacteria there was respectively a decrease and an increase in primary production values in the contaminated conditions. The increase observed for *Synechocystis* sp. (50 Zn =  $7.33 \mu\text{g C} \cdot 10^{-6} \text{cells}$ ) could be explained by the role of

Zn in carbonic anhydrase<sup>14,15</sup>, which could have led to a greater CO<sub>2</sub> fixation.

Chlorophyll *a* values for the 3 species are shown in Table 2. Despite the differences in pigment content between species, it is clear that controls had the highest values for both diatom and chlorophyte. *Synechocystis* sp. 100 Zn condition had the highest chlorophyll *a* value. The increase in chlorophyll *a* production has been reported for a variety of species submitted to high heavy metal concentrations<sup>16,17</sup>.

Differences in chlorophyll *a* content are responsible for the differences found in photosynthetic activity. This is shown in Table 3 and Fig. 1. *P. tricornutum* had a greater photosynthetic activity when cultivated under high Zn concentrations due to the lower values of the pigment in these conditions. The same happened to *Chlorella* sp. For *Synechocystis* sp. the reason for the increase in photosynthetic activity is due to the increase in primary production.

In the stressed conditions used, the increase in chlorophyll *a* seems to be the first sign for algae protection. However this does not reflect a higher photosynthetic activity as the increase in primary production does. Probably the increase in chlorophyll synthesis occurs as a compensatory mechanism in order to supply damages in photosynthetic structures. It is known that high heavy metals concentrations can damage chloroplasts<sup>18</sup>.

It could be speculated that protection mechanisms are used by stressed algae in order to keep the functioning of photosynthetic system. This, of course happens in heavy metal concentrations that are under the lethal ones. The survival of primary producers in such conditions, provided that some of them can accumulate metals<sup>19</sup>, can be dangerous in the sense of metal transfer through food chain in aquatic ecosystems.

TABLE 1: Primary production values (<sup>a</sup>ug C.l<sup>-1</sup>.h<sup>-1</sup>; <sup>b</sup>ug C.10<sup>-6</sup> cells) of the 3 species cultivated under different Zn concentrations

		Control	20 Zn	50 Zn	100 Zn
P.tricornutum	a	0.0130	0.0139	0.0198	0.0130
	b	3.07x10 <sup>-4</sup>	3.36x10 <sup>-4</sup>	3.85x10 <sup>-4</sup>	3.80x10 <sup>-4</sup>
Chlorella sp.	a	0.0677	0.0664	0.0620	0.0584
	b	1.35x10 <sup>-3</sup>	1.32x10 <sup>-3</sup>	1.1.x10 <sup>-3</sup>	1.01x10 <sup>-3</sup>
Synechocystis sp.	a	0.0422	0.0567	0.0595	0.0460
	b	4.95x10 <sup>-3</sup>	6.36x10 <sup>-3</sup>	7.33x10 <sup>-3</sup>	5.67x10 <sup>-3</sup>

TABLE 2: Chlorophyll a values (<sup>a</sup>ug.l<sup>-1</sup>; <sup>b</sup>ug.10<sup>-6</sup> cells) for the 3 species cultivated under different Zn concentrations

		Control	20 Zn	50 Zn	100 Zn
P.tricornutum	a	406.00	348.00	301.60	348.00
	b	0.422	0.362	0.235	0.337
Chlorella sp.	a	116.00	116.00	116.00	116.00
	b	0.164	0.140	0.120	0.133
Synechocystis sp.	a	1856.0	1624.0	1624.0	1624.0
	b	0.109	0.091	0.089	0.127

TABLE 3: photosynthetic activity values ( $\mu\text{g C} \cdot \mu\text{g Chl}_a^{-1} \cdot \text{h}^{-1}$ ) of the 3 species cultivated under different Zn concentrations

	Control	20 Zn	50 Zn	100 Zn
<i>P. tricornutum</i>	$8.00 \times 10^{-4}$	$9.19 \times 10^{-4}$	$10.8 \times 10^{-4}$	$11.2 \times 10^{-4}$
<i>Chlorella</i> sp.	$9.93 \times 10^{-3}$	$13.2 \times 10^{-3}$	$18.2 \times 10^{-3}$	$12.1 \times 10^{-3}$
<i>Synechocystis</i> sp.	$4.55 \times 10^{-3}$	$6.98 \times 10^{-3}$	$7.33 \times 10^{-3}$	$5.67 \times 10^{-3}$

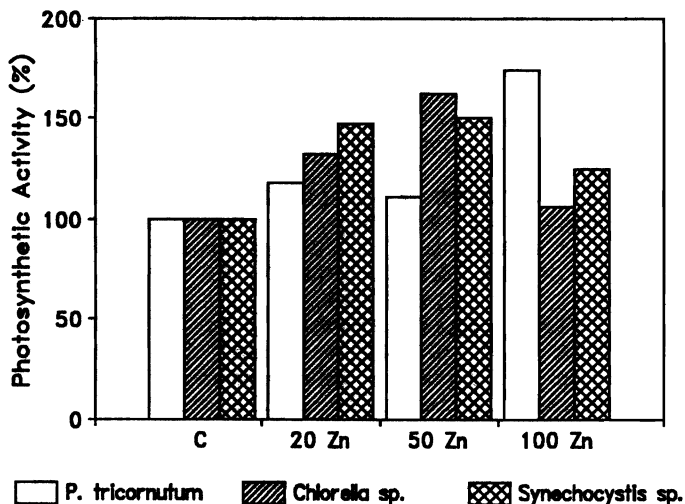


FIGURE 1: Photosynthetic activity of the 3 phytoplankton species cultivated under different Zn concentrations

**ACKNOWLEDGEMENTS:**

Thanks to Dr. Eduardo Penna-Franca and Rodolfo Paranhos for revision of this paper. This study was supported by CNPq (National Research Foundation).

**REFERENCES**

1. FORSTNER C.U. (1983) In FORSTNER C.U. (ed), Applied Environmental Geochemistry. Academic Press, London, 395-423.
2. PENNA-FRANCA E., PFEIFFER W.C., FIZMAN M. & LACERDA L.D. (1984) *Ciência e Cultura*, 36 (2): 215-219.
3. LIMA N.R.W., LACERDA L.D., PFEIFFER W.C. & FIZMAN M. (1986) *Environm. Technol. Letters*, 7: 453-460.
4. LACERDA L.D., PFEIFFER W.C. & FIZMAN M. (1987) *Sci. Total Environm.*, 65: 163-173.
5. REZENDE C.E., LACERDA L.D., ALVES M.A. & SOUZA C.M.M. (1987) VI Intern. Conf. Heavy Metals in Environm., 306-308.
6. ROBINSON N.J. (1989) *J. Appl. Phycol.*, 1: 5-18.
7. KAWAGUCHI S. & MAITA Y. (1990) *Bull. Environ. Contam.*, 45: 893-899.
8. GUILLARD R.R.L. (1975) In SMITH W.L., CHANLEY M.H. (eds.) *Culture of Marine Invertebrate Animals*. Plenum Publishing Corporation, New York, 29-59.
9. GUILLARD R.R.L. & LORENZEN C.J. (1972) *J. Phycol.*, 8: 10-14.
10. GIBSON C.E. & SMITH R.V. (1982) In CARR N.G. & WHITTON B.A. (eds.) *The Biology of Cyanobacteria*. Blackwell Scientific Publications, Great Britain, 463-490.
11. VIEIRA A.A.H. & AIDAR-ARAGÃO E. (1982) *Bolm. Inst. Oceanogr.*, 31 (1): 39-53.
12. BRAY G.A. (1960) *Anal. Biochem.*, 1: 279-285.
13. VOLLENWEIDER R.A. (1974) *A Manual for Measuring Primary Production in Aquatic Environments*. IBP Handbook No. 12, Blackwell Scientific Publications, Great Britain, 225 pp.
14. WIESSNER W. (1962) In LEWIN R.A. (ed.) *Physiology and Biochemistry of Algae*. Academic Press Inc., New York, 267-280.
15. SIMKISS K. (1979) *Endeavour, New Series*, 3 (1): 2-6.
16. GAETA S.A. (1987) *PhD. Thesis*. Univ. São Paulo, Inst. Oceanogr., 234.
17. TAKAMURA N., HATAKEYAMA S. & SUGAIA Y. (1990). *Jpn. J. Limnol.*, 51 (4): 225-235.
18. BOYLE T.P. (1984) In SHUBERT L.E. (ed.) *Algae as Ecological Indicators*. Academic Press Inc., 237-256.
19. JENSEN A., RYSTAD B. & MELSON S. (1974) *J. exp. mar. Biol. Ecol.*, 15: 145-157.

Accepted 24 May 1993