



## Effect of Industrial Effluent on the Growth of Marine Diatom, *Chaetoceros simplex* (Ostenfeld, 1901)

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**ABSTRACT:** The marine centric diatom, *Chaetoceros simplex* (Ostenfeld, 1901) was exposed to five different concentrations of industrial effluent for 96 hrs to investigate the effect on growth. The physico-chemical parameters viz. colour, odour, temperature, salinity, dissolved oxygen, turbidity, pH, alkalinity, hardness, ammonia, nitrite, nitrate, inorganic phosphate, total phosphorous, reactive silicate, calcium and magnesium were estimated in the effluent. The Ammonia ( $326 \mu\text{g. L}^{-1}$ ), Nitrite ( $19.53 \mu\text{g. L}^{-1}$ ) and Nitrate ( $471.4 \mu\text{g. L}^{-1}$ ) were observed at higher levels. About 50% of the cell density of *C. simplex* reached a lesser dilutions of effluent viz. 1:625 and 1:1250 than the control. The highest cell density ( $14.3 \times 10^4 \text{ cell ml}^{-1}$ ) was recorded in 1:10000 diluted effluent followed by control and the lowest cell density was observed in 1:625 diluted effluents. From the results, it is evidenced that the lower volume of effluent discharge into higher volume of water could not affect the growth rate of phytoplankton. It is more important that to reduce the effect of pollution and environmental sustainability. @JASEM

The primary producers are at the base of all ecosystems. The amazing property of algae and other autotrophs at incorporating energy and light into their metabolism and the food web is an unquestionable prerequisite of life. Beside this, plants in the coastal ecosystems constitute the living habitat for all other organisms by providing substrate for shelter and nursery areas for juvenile fishes etc. (Anderson, 1994; Pihl *et al.*, 1995; Eklund and Kautsky, 2003). The different demands upon the coastal ecosystem mean that it is important to develop good methods to assess the adverse impact on the organisms living there. Normally, toxicity tests on single species are used to establish the toxicity of chemicals and complex waste waters, inhibitory actions of xenobiotics on growth and other physiological functions of algae, e.g. by industrial outlets will affect the whole aquatic ecosystem. Tests of growth inhibition of microalgae are therefore frequently used to characterise industrial effluents (Thomas *et al.*, 1980; Geis *et al.*, 2000; Eklund and Kautsky, 2003).

Rapid industrialization in India has resulted in the substantial increase in the liquid waste (spent wash or effluent), which is traditionally discharged into open land or into nearby natural water, causing a number of environmental problems including threat to plants and animal lives and also creating problems such as surface water logging, ground water contamination and salinizing good quality land due to presence of high amount salt contents (Ramona *et al.*, 2001). Nitrogen and phosphorus contained in agricultural effluents and industrial discharges are the main cause of eutrophication. Treated wastewater usually still contains ions such as nitrate, nitrite, ammonium and phosphate, which provide nutrients for algal growth in seas and lakes, resulting in nuisance growths. In the present study, *C. simplex* was selected as a model species of marine diatoms to investigate the toxic effects of industrial effluent on marine diatoms at the

population level (effect on growth) and to explore whether they can be used as a valuable index in assessing the potential risk to phytoplankton communities.

### MATERIALS AND METHODS:

**Test Species:** The unialgal culture of marine diatom, *C. simplex* was accomplished in Phytoplankton Culture Laboratory, CAS in Marine Biology, Faculty of Marine Sciences, Annamalai University, Parangipettai. The seawater was collected at 10 km away from shore of the Parangipettai coast. The seawater was kept in a dark room for three months and the aged seawater was filtered through 0.2  $\mu$  membrane filter. The filtered seawater was amended with f/2 nutrients (Guillard, 1975), sterilized by autoclaving at 121°C for 15 minutes and used as medium.

**Physico-chemical Parameters:** The industrial effluent was collected from discharging site of Cuddalore industrial area, Tamil Nadu. The physico-chemical parameter, temperature was determined by Celsius thermometer, pH by Electronic pH meter (ELICO, Model LI 120), Salinity by Hand Refractometer (ERMA, Japan), Turbidity by Digital Turbidity meter (ELICO, Model 331), Dissolved Oxygen by Winkler's method, Alkalinity, Ammonia, Nitrite, Nitrate, Total Phosphate, Inorganic Phosphate, Reactive Silicate, Calcium and Magnesium by the method of Strickland and Parsons, (1972). One-Way ANOVA and Duncan's multiple range tests were performed for growth rate in SPSS 10.0 software.

**Experimental Design:** A subculture was prepared in 5 litre culture flask with three litres of f/2 media and it was allowed to reach exponential phase for 4 days. Experiments were conducted in duplicate to estimate the effect of industrial effluent by exposing the marine diatom, *C. simplex* (initial cell density  $1.7 \pm 0.28 \times 10^4 \text{ cells. ml}^{-1}$ ) with the following five

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different concentrations of the industrial effluent for 96 hours: 1) 1ml effluent in 10000 ml water (1:10000); 2) 1 ml effluent in 5000 ml water (1:5000); 3) 1 ml effluent in 2500 ml water (1:2500); 4) 1 ml effluent in 1250 ml water (1:1250); 5) 1 ml effluent in 625 ml water (1:625).

The following parameters were maintained during the experiment: Temperature:  $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$ ; Salinity: 30 psu; Light:  $4500 \pm 500$  Lux

**Cell density and Growth Rate:** The cell growth was monitored at the beginning and end of the experiment by measuring cell numbers by manual counting under the binocular light microscope. Growth rates were calculated as  $\mu$ .  $\text{day}^{-1}$  according to the following formula:

$$\mu = \frac{N_1 - N_0}{t}$$

where,  $N_0$  and  $N_1$  represent cell density at the start and the end of the growth period, and  $t$  is the time between measurements (in days).

## RESULTS AND DISCUSSION

In the experiment, some physico-chemical parameters were shown at elevated levels. The colour was yellowish green and the odour was obnoxious. The ammonia ( $326 \mu\text{g. L}^{-1}$ ), nitrite ( $19.53 \mu\text{g. L}^{-1}$ ) and nitrate ( $471.4 \mu\text{g. L}^{-1}$ ) were observed at elevated levels (Table 1). The cell density was significantly decreased with the increasing concentration (decreasing dilutions) of the effluent. However, the growth was slightly enhanced with the minimum concentration (higher dilution) of the effluent. The lesser dilutions of effluent viz. 1:625 and 1:1250 were highly affected the multiplication of *C. simplex* about 50% of the cell density only reached than the control. The highest cell density ( $14.3 \times 10^4 \text{ cell ml}^{-1}$ ) was recorded in 1:10000 diluted effluent followed by control and the lowest cell density was observed in 1:625 diluted effluents (Fig. 1).

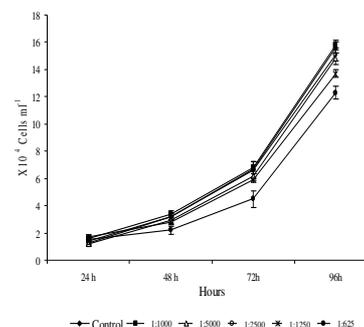
**Table 1:** Physico-chemical parameters of the effluent

S. No.	Parameter	Range
1	Colour	Yellowish green
2	Odour	Obnoxious
3	Turbidity	10 NTU
4	Temperature	$27^{\circ}\text{C}$
5	Salinity	5 ‰
6	pH	7.5
7	Dissolved oxygen	$2.5 \text{ ml. L}^{-1}$
8	Alkalinity	0
9	Ammonia	$326 \mu\text{g. L}^{-1}$
10	Nitrite	$19.53 \mu\text{g. L}^{-1}$
11	Nitrate	$471.4 \mu\text{g. L}^{-1}$
12	Total Phosphate	$47.66 \mu\text{g. L}^{-1}$
13	Inorganic Phosphate	$23.79 \mu\text{g. L}^{-1}$
14	Reactive Silicate	$60.2 \mu\text{g. L}^{-1}$
15	Calcium	$1.9 \text{ mg. L}^{-1}$
16	Magnesium	$1.73 \text{ mg. L}^{-1}$

The average daily specific growth rates were significantly different between the effluent concentrations at 0.05% level. It was decreased with

decreasing dilutions of the effluent. The highest growth rate was observed in 1:10000 diluted effluent followed by control, 1:5000, 1:2500, 1:1250 and lowest growth rate was in 1:625 dilutions (Fig. 2).

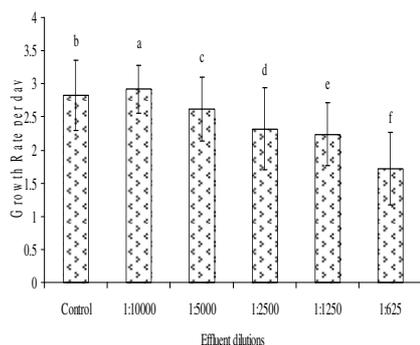
In the present study, the ammonia ( $326 \mu\text{g. L}^{-1}$ ) concentration in the effluent was found similar as reported by Livingston *et al.* (2002) and the effluent ammonia concentration was 190 to  $430 \mu\text{g. L}^{-1}$ . Livingston *et al.* (2002) indicated Nitrogen was one of the chief limiting nutrients to phytoplankton through nutrient limitation experiments. But the high concentrations of ammonia, nitrite and nitrate concentrations were affected the general multiplication and physiology of phytoplankton with pH conditions of the culture (Kallqvist and Svenson, 2003). The effluent tested in the present study has higher values of ammonia, nitrite, nitrate and the growth of *C. simplex* was slightly enhanced with lower concentration and inhibited at higher concentrations of the effluent.



**Fig.1:** Cell density (Mean  $\pm$  SD) in different dilutions of the industrial effluent

Kallqvist and Svenson (2003) have been reported that the growth rate inhibition of microalgae *Nephroselmis pyriformis* related to the concentrations of ammonia in the medium. Livingston *et al.* (2002) also studied the laboratory microcosms and field mesocosms to determine whether or not ammonia had a toxic effect on plankton assemblages and to analyze possible effects of mill effluents on ammonia impacts. The microcosm results indicated that ammonia had a stimulatory effect on *S. costatum* at mean concentrations of  $0.06 \text{ mg l}^{-1}$  with negative effects of ammonia occurring within a range of  $0.1-0.24 \text{ mg l}^{-1}$  and major impacts at concentrations  $>0.46 \text{ mg l}^{-1}$ . The mesocosm experiments with ammonia indicated stimulatory effects from  $0.11$  to  $0.14 \text{ mg l}^{-1}$  and inhibition of phytoplankton growth beyond  $0.20 \text{ mg l}^{-1}$ . The difference between stimulatory effects and inhibition of ammonia on *S. costatum* and phytoplankton assemblages was relatively small. The same observations were recorded in the present experiment on *C. simplex* with the industrial effluent. Where the growth and growth rate of *C. simplex* was enhanced with the higher dilution (lower concentration) of the effluent and the

growth was inhibited in the higher concentrations of the effluent (1:625 dilutions).



**Fig.2:** Average Daily Specific Growth Rate of *C. simplex* (Mean ± SD) with different dilutions of the industrial effluent

The lower concentration of the effluent was enhancing the growth of phytoplankton and will create the blooming problem if the species stimulated by the effluent is harmful. On the other hand, the higher concentration of the effluent was inhibiting the growth of phytoplankton and will reduce the primary production, availability of food source to zooplankton and other larval forms. Still the toxicity study on phytoplankton is so lacking which need more studies on the phytoplankton to develop the sea water quality criteria.

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