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Preliminary report on the tolerance and growth of micro algae in the sludge from hypochlorite manufacturing industry and possibility of using these micro algae for remediation.

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Abstract

The ETP sludge of a hypochlorite manufacturing industry can be a potent pollutant of an aquatic environment. The hyposludge is highly alkaline with high TDS. It is milky white with sodium hypochlorite as a major constituent. The present work attempts to identify micro algae which can tolerate and grow in the hyposludge, Preliminary laboratory screening revealed that *Chlorococcum humicola* and *Chroococcus turgidus* showed a significant growth. These two micro algal species can be employed for remediation of hyposludge after systematic lab trials followed by pilot scale studies.

Keywords: *Chroococcusturgidus*, *Chlorococcumhumicola*, Phycoremediation, Hypo sludge

Introduction

Pollution is the introduction of a contaminant into the environment.Water pollution is the introduction of chemical, biological and physical matter into large bodies of water that degrade the quality of life that lives in it and consumes it. The causes of water pollution may be due to direct (Point source) and indirect (Non-point source) contaminant sources. The direct sources are effluent outputs from refineries, factories and waste treatment plants.

Worldwide contaminated and polluted water now kills more people than all forms of violence including wars, according to a United Nations report called “Sick Water?” (UNEP, 2010) released on March 22, 2010 on World Water Day that calls for turning unsanitary wastewater into an environmentally safe economic resource.

Microalgae being the producers of aquatic environment play a significant role in the wellbeing of the atmosphere. Micro algal research has many advantages as they have the ability to convert CO_2 to useful materials through photosynthesis. Microalgae are used as single cell protein as food supplement. They are used as human nutritional supplements, as animal feed additives, in aquaculture, and in cosmetics. It is used by astronauts during space travel. Microalgae are tapped as a source for useful secondary metabolites. Recently, algae have been associated with regulating environmental pollution. These can help in removing pollutants from the surroundings, restoring contaminated sites and preventing further pollution. Lately, a great deal of interest has been centered on algae as potential candidates for bioremediation of polluted water bodies (Hassett et al, 1981 and Fernandez-pinas et al, 1991). Algae fit well with this *'green movement'* of maintaining nature's harmony through microorganisms (Atlas, 1995).

Phycoremediation, as defined in a broad sense, is the use of macroalgae or microalgae for the removal or biotransformation of pollutants, including nutrients and xenobiotics from wastewater and CO2 from waste air (Olgu’in, 2003). Phycoremediation is a novel technique that uses algae to clean up contaminated soil and water. The first ever phycoremediation plant with *Chroococcusturgidus*has been working from September 2006 at SNAP industry very successfully (Sivasubramanian *et al*, 2009).

Industries which are manufacturing calcium hypochlorite ($Ca(ClO)_2$) use chlorine and Milk of Lime in their Chlor alkali plant. In the process of manufacturing impurities like calcium carbonate, sand and silica settle down in the settler. Periodical draining of the impurities, leads to accumulation of sludge with high quantity of calcium hypo chlorite and calcium chloride. The process of washing of sludge leads to the increase in the effluent quantity.The wastewater produced by these industries is a serious environmental hazard because it contains diverse chemical compounds like calcium carbonte, calcium chloride, calcium hydroxide, magnesium hydroxide, etc.

Calciumhypochlorite is widely used for water treatment and as a bleaching agent (bleaching powder). It is considered to be relatively stable and has greater available chlorine than sodium hypochlorite (liquid bleach). It is also used in swimming pools and cooling towers of industries to avoid algae and mosses. Calcium Hypochlorite is used to manufacture of Chloroform and bleaching sugarcane juice before sugar crystallization. The usage of highly toxic chemicals in the industry makes the generated wastewater non-ecofriendly. The sludge is a concentrated form of the wastewater. It is highly toxic even in small quantities. The present study is aimed at identifying suitable micro algal strains which can be used in the phycoremediation of effluent sludge of hypochlorite manufacturing industries. .

Materials and Methods

Hyposludge

Hyposludge has been obtained from a hypochlorite manufacturing industry located in Madhya pradesh, India. The sludge is milky white in colour with high TDS and highly alkaline in nature. The sludge has high quantity of calcium

carbonate and considerable quantity of calcium chloride, calcium hydroxide and magnesium hydroxide and traces of Iron. (Table 1)

Table 1. Chemical composition of hyposludge

PARAMETERS		% (W/W)
Hydroxides	**$Ca(OH)_2$**	**4.5-6.5**
	$Mg(OH)_2$	**7-10**
Chlorides	**$CaCl_2$**	**7-10**
Carbonates	**$CaCO_3$**	**25-30**
Acid Insoluble		**40-45**
Sulphates	**Na_2SO_4**	**1-2**
Iron	**Fe**	**0.001 -0.005**
pH		**10.5-11.5**

Micro algae

The microalgae employed in the present investigation were obtained from Vivekananda Institute of Algal Technology (VIAT) culture collection, Chennai. Eight species of microalgae used are listed below:

1. *Chroococcus turgidus*
2. *Chlamydomonas pertusa*
3. *Chlorella vulgaris*
4. *Chlorococcum turgidus*
5. *Dactylococcopsis raphidioides*
6. *Desmococcus olivaceous*
7. SNAP isolate 1
8. SNAP isolate 2

Screening of micro algae for tolerance and growth

Two different effluent sludge concentrations namely 2.5% and 5 % were prepared in test tubes with culture medium as a diluting agent. Centrifuged micro algal pellets of the above mentioned species were added separately to the effluent

concentrations. The test tubes were incubated for seven days after observing the initial pH, conductivity and cell count. The growth rates were calculated.

Results and Discussion

The effluent analysis showed that the sludge was alkaline in nature with high TDS. It makes the sludge highly toxic in nature. The micro algal growth in the sludge was studied by diluting it with culture medium in two different concentrations (2.5% and 5%). The growth rates were calculated after seven days of incubation. All the species inoculated in 5% concentration hardly showed any growth. This may be due to presence of chemicals like Calcium hypochlorite and Calcium Chloride in the sludge. In 2.5% concentration only four microalgae showed positive growth rate *Chroococcus turgidus*, *Chlorococcum humicola*, SNAP isolate 1 and SNAP isolate 2 (**Table 2**) were the microalgae which showed growth in the sludge . *Chlorococcum* sp showed a better growth rate than the other three species.

Table 2 Growth of micro algae in hyposludge from hypochlorite manufacturing industry

S.No	Microalgae	Growth	
		5% sludge	2.5% sludge
1	Control	0	0
2	*Chroococcus turgidus*	0	0.023
3	*Chlamydomonas pertusa*	0	0
4	*Chlorella vulgaris*	0	0
5	*Chlorococcum humicola*	0	0.040
6	*Dactylococcopsis raphidioides*	0	0
7	*Desmococcus olivaceous*	0	0
8	SNAP Isolate 1	0	0.024
9	SNAP Isolate 2	0	0.018

The growth in 2.5% concentration maybe due to very low concentration of sludge and presence of carbonates (Calcium Carbonate). The presence of Iron may also help micro algal growth. Iron is also responsible for enhancing phytoplankton

biomass in oceanic waters (Lin, Z Y *et al*, 2008). Iron is responsible for increase in photosynthesis by increasing the chlorophyll content. Being an autotroph, CO_2helps algae for good growth. CO_2 is the readily absorbed form of carbon though algae can also utilize carbonate (CO_3^-) and bicarbonate (HCO_3^-). CO_2 is transported across the plasma membrane and algae cells incorporate it in the form of HCO_3^-. The enzyme carbonic anhydrase is responsible for the conversion of HCO_3^- to CO_2 (Badger, M R *et al.*, 1994 and Emma Huertas, *et al.*, 2000). This enzyme also helps in the inorganic carbon uptake at alkaline pH and low CO_2 concentration in the growth culture (Moroney *et al.*, 1985).

Conclusion

The present study in laboratory has shown that two micro algae could tolerate and grow in hyposludge. *Chlorococcum humicola* and *Chroococcus turgidus* showed a significant growth. These two micro algal species can be employed for remediation of hyposludge after systematic lab trials followed by pilot scale studies.

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