



# Impact Of Effluent Of A Chlor-Alkali Industry On The Atpase Activity Of A Cyanobacterium, *Westiellopsis Prolifica*, Janet Under Laboratory Controlled Conditions.

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## Highlights:

- The effluent of the Chlor-alkali industry is deadly toxic and showed drastic effect at higher concentrations.
- The effluent contained significant amount of mercury both elemental and inorganic.
- The exposed alga showed dual behavior to the effluent at different concentrations.
- The ATPase activity showed stimulatory effect at sub-lethal concentrations of the effluent.
- The effluent showed inhibitory effect at higher concentrations of the effluent.
- The effluent influenced the energy budget by way of interfering in the energy trapping and energy transfer mechanism of the exposed alga.

## Abstract

The present study was aimed to analyze the Physico-chemical nature of effluent of Chlor-alkali industry collected from the effluent canal and its impact on a BGA. To study the impact of effluent on the ATPase activity an important enzyme related to energy management of the effluent exposed alga. The cyanobacterium (BGA) showed dual behavior towards the effluent toxicant indicating stimulation at sub-lethal concentrations and inhibition at higher concentrations of the effluent toxicant. During recovery studies interesting results were obtained. Significant recovery at sub-lethal concentration of the toxicant and insignificant recovery at higher concentrations of the mercury contained toxicant during recovery period indicated dual behavior of the toxicant. Significant variations were observed in the ATPase activity of the control and effluent exposed alga. The control alga showed normal ATPase activity with the increase in exposure period. At sub-lethal / maximum allowable concentration of effluent, the exposed alga showed increase in ATPase activity compared to ATPase activity of the control alga indicating higher growth. Whereas, at higher concentration of the effluent, depletion in ATPase activity was noted in the effluent exposed algal sets. Stimulation of ATPase activity in the effluent exposed alga at sub-lethal concentrations of mercury contained effluent and inhibition in ATPase activity at higher doses of mercury contained effluent indicated the possible interference of mercury in exposed algal physiological processes of energy trapping mechanism, oxygen evolution and primary production and energy utilization. During recovery studies interesting results were obtained. Significant recovery at sub-lethal concentration of the toxicant exposed algal ATPase activity and insignificant recovery at higher concentrations of the mercury contained toxicant during recovery period indicated dual behavior and impact of the toxicant.

**Keywords:** Chlor-alkali industry, effluent, mercury, Blue-green alga, ATPase activity.

## Introduction

It is an accepted fact that all industries produce pollutants. The pollutants may be gases (air pollutants) or liquids (effluent- water pollutants) or solids (Solid waste- Land pollutants) are released / produced from the industries during the processing of raw material and production of Products of Interest (POI) as unwanted end products or by-products or intermediaries as wastes. The origins of these pollutants are from the industries causing pollution of surrounding air, water bodies and landmasses. Pollution whose source originates directly from the industry and causes severe health hazards is known as industrial pollution. All the industries use raw materials obtained from nature and in the industrial processing phase convert the chemicals into different forms and varieties of chemicals are produced. Out of which few produced chemicals are of our interest and many more chemicals are not required by us and once available in the environment cause health hazard. These unwanted chemicals are many types with different configurations and once discharged into the natural environment will affect both plant and animal life and most important the health of the ecosystem will deteriorate with time. The impact of these unwanted chemicals which we call as waste may cause acute or chronic effects. The industry and its waste is the most important source of pollution of the environment. After the great industrial revolution, manufacturing and technology made advances, which resulted in more factories and more industries appearing on the environmental screen. The smoke released from the factories are emitted into the air through chimneys of different heights affect the plants and animals. The liquid effluents produced by the industries are discharged into fresh water bodies causing serious impact on aquatic life. It was also reported that the effluents after discharge in to the water bodies contaminate the ground water and cause serious problems for all plant and animal life. The solid wastes produced by the industries are generally dumped at near by places causing land pollution. When these solid wastes were transported to distances and thrown carelessly at different sites cause serious land pollution. Industrial pollution was also pointed out as a major factor in loss of biodiversity, loss of medicinal plants, loss of endangered species, wildlife extinction and eventually the air pollutants become a cause for global warming. All these air pollutants, water pollutants and land pollutants move to distances impacting the plants and animals inhabiting in different environmental segments.

The effluent of the Chlor-alkali industry contains a significant amount of mercury as reported by many authors working on different industries and causing serious threat to the environment, aquatic plant and animal life and finally human beings are also affected. Mercury (Hg) is a naturally occurring unique silvery metallic element with special property of being liquid at room temperature (Norrby, 1991; Tangahu *et al.*, 2011). Both anthropogenic activities and natural processes cause its release into different spheres of the environment resulting in severe adverse impacts was reported by Sundseth *et al.* (2015). Increased anthropogenic discharge of mercury leads to disturbance in its natural biogeochemical cycle which results in to unenviable diseases and hazardous health effects (Zhang *et al.*, 2014). Padhy and Panigrahi (2018a, b) reported the effects of heavy metals like Cd & Pb on the photosynthetic efficiency, pigment content, growth and nitrogen fixation of a cyanobacterium under experimental conditions. Sahu (2017a, b) reported the effects of mercury contained wastes on cyanobacterium and distribution of mercury contained waste on a cyanobacterium and possible reclamation. The authors are of the opinion (Prusti and Panigrahi, 2017a, b) that regular intake of mercury by the plant and its enrichment in the body led to increase in mercury body burden which interfered in all metabolic activities and finally killed the plant. The present study was planned to study the impact mercury contained effluent on the ATPase activity of a blue-green alga, *Westiellopsis prolifica*, Janet in laboratory conditions.

## Materials & Methods

**Location of the industry:** M/S Jayashree Chemicals Pvt. Ltd., was situated at Ganjam, on the Bank of Rushikulya estuary about 1.5 km. Away from the sea, Bay of Bengal, on the East and 30 km. North of Berhampur city on the south-eastern side of India at 84<sup>0</sup>53'E longitude and 19<sup>0</sup>16'N latitude with a mercury cell electrolysis process of manufacturing caustic soda..

**Toxicant:** Effluent of the Chlor-alkali industry collected from effluent canal.

**Test organism:** *Westiellopsis prolifica*, Janet; Family: Stigonemataceae.

Physico-chemical analyses of effluent and solid waste and crop field soil samples of nearby crop fields were conducted periodically by following the procedure of APHA (1998), standardized field analysis kit. Effluent samples were brought in glass containers and stored in cold room for use in laboratory experimental work. The alga was grown in Allen & Arnon's culture medium (1955) with trace elements (Fogg, 1949), modified by Patnaik (1964). The cultures were maintained in culture racks at an illumination of 2200±200Lux and temperature was maintained at 26±2°C. The culture flasks were hand shaken twice daily to avoid clumping and adherence of the algal filaments to the walls of the culture vessels. Total ATPase activity was determined by measuring the amount of i. p. (inorganic phosphate) released when ATP was converted to

ADP by following the molybdenum blue method as described by Martineck (1970). The obtained data was statistically analyzed to test the level of significance.

## Results

Table-1: Physico-chemical analysis of the effluent collected from a select point near the effluent storage tank of the industry. Data are the mean of 4 estimations  $\pm$  standard deviation.

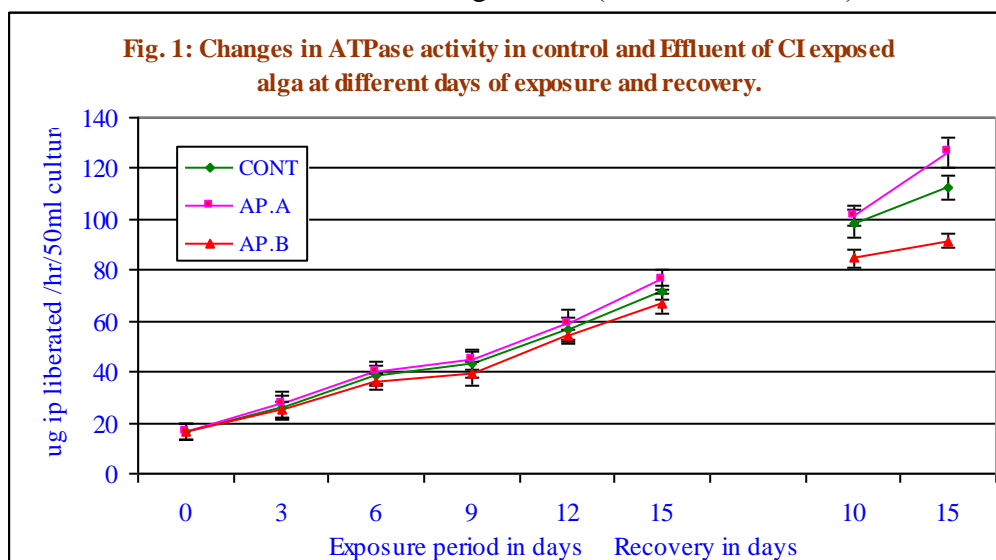
Sl. No	Parameters	Effluent (mg / l)	
		storage tank	From discharge point
1	Temperature ( $^{\circ}$ C)	29.2 $\pm$ 1.5	30.2 $\pm$ 1.5
2	pH	9.2 $\pm$ 0.3	9.3 $\pm$ 0.2
3	Mercury	0.49 $\pm$ 0.06	1.08 $\pm$ 0.32

No significant variation in temperature and pH was marked in the effluent collected from two spots, the solid waste and extract prepared from solid waste. The pH of the effluent was highly alkaline. The effluent and solid waste analysis indicated presence of significant amount of mercury much higher than the prescribed limit (Table-1).

Table- 2: Lethal concentration values obtained from toxicity regression analysis curve ( $r=0.987$ ; P (Level of significance)  $\geq 0.01$ ; MAC=Maximum Allowable Concentration, MPS= Maximum Percent Survival)

Sl. No	Lethal concentration values (LC)	Effluent concentration. ml/50ml culture (v/v)	Percent survival values (PS)
1	MAC (LC <sub>00</sub> ) AP.A	0.092	MPS (PS <sub>100</sub> )
2	LC <sub>10</sub> AP.B	0.11	PS <sub>90</sub>
3	LC <sub>50</sub>	0.21	PS <sub>50</sub>

The LC<sub>00</sub>, LC<sub>10</sub>, LC<sub>50</sub>, LC<sub>90</sub> and LC<sub>100</sub> values of the chlor-alkali industry effluent were and deduced from toxicity testing. The maximum allowable concentration / dose for the tested alga was found to be 0.092ml of the effluent / 50ml culture for 15days of exposure. The lethal concentration LC<sub>10</sub> or PS<sub>90</sub> value found to be 0.11ml of the effluent / 50ml culture solution where 10% death and 90% survival was noted. The lethal concentration (LC<sub>50</sub>) value found to be 0.21ml of the effluent / 50ml culture solution where 50% death was noted. The lethal concentration (LC<sub>90</sub>) value found to be 0.32ml of the effluent / 50ml culture solution where 90% death was noted. At 0.41ml effluent solution / 50 ml culture, 100% death of the alga was observed. From the toxicity data we can clued that more than 0.41ml/50ml culture is deadly toxic where the exposed alga is unable to survive where percent survival was zero (Table-2). Significant variations in ATPase activity of control and Chlor-alkali industry effluent exposed cyanobacterium were marked and have been shown in Fig.1. The ATPase activity showed gradual increase with the increase in biomass and with the increase in exposure period in control and effluent exposed alga to different concentrations of the effluent. The ATPase activity increased from 16.5  $\pm$  2.8 $\mu$ g ip liberated /hr / 50 ml culture on 0 day of exposure to 72.2  $\pm$  4.6 $\mu$ g ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of exposure in the control set. The control alga when transferred to normal culture medium the enzyme activity increased significantly to 113.2  $\pm$  5.2 $\mu$ g ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of recovery. The increase in the enzyme activity was positive and the correlation coefficient value was significant ( $r = 0.991$ ;  $P \geq 0.001$ ).



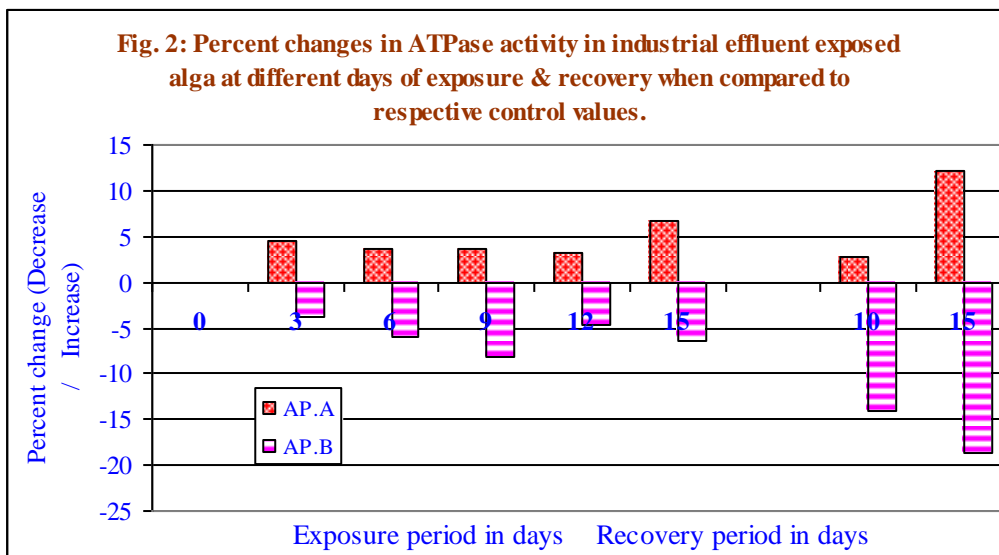


TABLE-I: ANOVA Single factor for total ATPase activity data analysis

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	5975.109	5	1195.022	186.2694	6.11E-11	3.105875
Within Groups	76.98667	12	6.415556			
Total	6052.096	17				

There are non significant differences observed between rows and columns.

The ATPase activity increased from  $16.5 \pm 2.8 \mu\text{g}$  ip liberated /hr / 50 ml culture on 0 day of exposure to  $44.5 \pm 3.9 \mu\text{g}$  ip liberated /hr / 50 ml culture on 9<sup>th</sup> day of exposure in the effluent exposed set at conc. AP.A (MAC value) set. The enzyme activity value further increased to  $76.2 \pm 4.1 \mu\text{g}$  ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of exposure in the conc. AP.A set. The exposed alga when transferred to toxicant free normal culture medium the enzyme activity increased significantly to  $126.4 \pm 5.9 \mu\text{g}$  ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of exposure (Fig.1). The increase in the enzyme activity was positive and the correlation coefficient value was significant ( $r = 0.988$ ;  $P \geq 0.01$ ). The ATPase activity increased from  $16.5 \pm 2.8 \mu\text{g}$  ip liberated /hr / 50 ml culture on 0 day of exposure to  $39.4 \pm 4.4 \mu\text{g}$  ip liberated /hr / 50 ml culture on 9<sup>th</sup> day of exposure in the effluent exposed set at conc. AP.B (LC<sub>10</sub> value) set. The enzyme activity value further increased to  $66.8 \pm 3.6 \mu\text{g}$  ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of exposure in the conc. AP.B set. The exposed alga when transferred to toxicant free normal culture medium the enzyme activity increased significantly to  $91.5 \pm 2.9 \mu\text{g}$  ip liberated /hr / 50 ml culture on 15<sup>th</sup> day of exposure. The increase in the enzyme activity was positive and the correlation coefficient value was significant ( $r = 0.988$ ;  $P \geq 0.01$ ). From the data it is very clear that the effluent containing mercury has no significant effect on the enzyme activity at maximum allowable concentration and at LC<sub>10</sub> of the effluent. It was also observed that there was no significant difference in the enzyme activity between effluent concentrations and control at different days of exposure, as the obtained values were well within the standard deviation range. The regression analysis indicated no significant (NS) difference at all exposure periods except on where 9<sup>th</sup> and 12<sup>th</sup> day regression value was significant but 0.1 and 0.11 level. The analysis variance ratio test indicated similar trend in both the rows and columns (Table-I). The calculated percent change values of course indicated increase in the enzyme activity at conc. AP. A (MAC value) and decrease in enzyme activity at conc. AP.B effluent concentration (Fig. 2). At sub-lethal concentration of the effluent maximum 6.7% increase in the enzyme activity was noted on 15<sup>th</sup> day of exposure and LC<sub>10</sub> effluent concentration 6.4% decrease on the enzyme activity was observed. From toxicological stand point the increase at lower concentration and decrease in the parameters studied at higher concentration carries a meaning and can reflect the status of the effluent pollutant. The death of the exposed system depends upon its age, tolerance, resistance, physiological ability to withstand stress and biochemical mechanism of the organism to defend stress.

## Discussion

Mercury gets into the environment through a process known as mercury cycle. It was observed that the total mercury estimated in the effluent was high and contained mainly three types of mercury. The death of the exposed system depends upon its age, tolerance, resistance, physiological ability to withstand stress and biochemical mechanism of the organism to defend stress. The most significant source of mercury availability in the environment was mostly due to the Chlor-alkali industries (Hartung & Dinman, 1974; Bothner & Carpenter, 1973 and Fimreite, 1970). The above observations is a clear indication that status and intensity of mercury poisoning is improving by way of depletion of mercury level in air, water and land mass and the environment is moving towards better and favorable, where all types' plants both micro and macro can grow and survive. Heavy metal ATPases (HMAs) are P1B-type ATPases that play a significant role in metal trafficking in plants (Huang *et al.*, 2020a). Heavy metal ATPases (HMAs) of the P1Btype have been linked to the transport of a variety of necessary and potentially hazardous metals across cell membranes. The heavy metal can alter the activity of enzymes either by binding to their functional groups such as sulphidryl, carboxyl and imidazol or by displacing the metal associated with the enzyme (Viarengo, 1985). ATPase enzymes are -SH rich enzyme which play a pivotal role in teleost intestine and gill physiological functions such as salt- and osmoregulation and acid-base balance. The plant plasma membrane (PM) can be regarded as the first structure that is a target for heavy-metal toxicity. An increase in permeability related to membrane damage is observed in plants that have been subjected to heavy-metal stress (Demidchik *et al.*, 1997, 2001; Murphy and Taiz, 1997; Murphy *et al.*, 1999). It is well known that metal ions are easily bound to both the sulphhydryl groups of proteins and hydroxyl part of phospholipids (Devi and Prasad, 1999). They can also replace the Ca<sup>2+</sup> ions at essential sites of cell membranes (Breckle and Kahle, 1991). All these events result in disruption of membrane integrity and ionic homeostasis of cells. Thus, tolerance may involve the protection of plasma membrane integrity against heavy-metal damage and maintaining ionic balance (Janicka-Russak *et al.*, 2012). Mechanisms of Cu<sup>2+</sup>-induced H<sup>+</sup>-ATPase suppression have been investigated in vitro (Serrano *et al.*, 1985; Serrano, 1990). According to these authors, Cu<sup>2+</sup> ions are one of the most powerful inhibitors of PM H<sup>+</sup>-ATPase, producing half-maximal inhibition at 2–5 μM and complete inhibition at 10–20 μM. The inhibitory effect of Cu<sup>2+</sup> seems to be related to its interaction with sulphhydryl groups at the active site of the ATPase. The inhibitory effect of Cu<sup>2+</sup> seems to be related to its interaction with sulphhydryl groups at the active site of the ATPase. The effect of metals on PM H<sup>+</sup>-ATPase activity depends on time of exposure of plants to heavy metals, the type and concentration of heavy metal and the plant species (Janicka-Russak *et al.*, 2012). Osmoregulation is the ability to actively maintain osmotic concentrations in extracellular fluids, in spite of the osmolarity (salinity) of the surrounding environment. The assessment of ATPase activity may therefore be used as an early warning signal of metal-induced damage to the osmoregulatory and acid-based regulatory system in osmoregulatory organs such as gills, kidney and intestine (Monteiro *et al.*, 2005; Atli and Canli, 2007). Heavy metals stimulate enzyme activity via the transformation process of maternal or embryonic tissues and display in the form of natural growth variations Koshan *et al.*, 2021). In 1986, Kramer *et al.* (1986) observed that among other heavy metals, Hg<sup>2+</sup> ranked the highest in their inhibition of the Na<sup>+</sup>/K<sup>+</sup>-ATPase. Therefore, their finding showed that mercury toxicity on sodium pump is of significance and deserved critical attention. In the present study it was observed that mercury contained effluent stimulated the enzyme activity in blue-green alga and inhibited the enzyme activity at higher concentrations of the effluent waste and higher exposure period, ultimately killing the organism. It is a fact that at higher mercury contained effluent waste exposure the alga died because of interference of the heavy metal, mercury in growth, physiological process and biochemical processes. Heavy metals stimulate enzyme activity via the transformation process of maternal or embryonic tissues and display in the form of natural growth variations Koshan *et al.*, 2021). In 1986, Kramer *et al.* (1986) observed that among other heavy metals, Hg<sup>2+</sup> ranked the highest in their inhibition of the Na<sup>+</sup>/K<sup>+</sup>-ATPase. Therefore, their finding showed that mercury toxicity on sodium pump is of significance and deserved critical attention.

In the present study we marked that the Chlor-alkali industry effluent showed dual behavior like stimulation at sub-lethal concentration and inhibition at higher concentration of the effluent containing significant amount of mercury. The exposure period also plays a crucial role in toxicity study. At lower exposure period, the exposed system might behave in a different way compared to the system's behavior at higher exposure period. Our findings agree with the reports of Rath (1984) not authenticated earlier but confirmed now and we agree with the suggestions and observed trends of Sahu (1987). In case of heavy metal, copper, the growth regulation or stimulation can be explained, as this particular heavy metal comes under essential elements added in the culture nutrients. Spencer and Nichols (1983) reported further that inorganic forms are active only when these are present in free ionic state. Our finding is well supported by the reports of Prasad and Prasad (1982) as they observed stimulation of the algal growth at low concentration of

Cadmium, Lead and Nickel. O'Kelley (1974) reported neither Cd / Pb / Ni have been reported to be essential micro-nutrients required for the growth and the development of algae nor the pure solution of these heavy metals are expected to act as growth regulator for the exposed system. Gadd & Griffith (1978) reported that biological activity of heavy metal ions is markedly affected by the presence or absence of other ions and other essential or non essential chemicals / debris. Mishra *et al.*, (2013); Shaw *et al.*, (1989) and Sahu *et al.* (1986) and many other authors have found that heavy metals cause prolongation of lag phase more or less in proportion to doses, followed by normal growth (Zingmark and Miller, 1975). We agree with the common explanation for stimulation at lower concentration is probably that the medium is modified during the first part of the experiment either by exudation from living cells or non availability of binding sites or non penetration of the heavy metals in to the cell due non permeability of the membrane or by leaching of chemicals from dead cells or decomposed debris of dead cells to render the heavy metal less toxic by some sort of chelation (Shaw, 1987) or any other special mechanism yet to be confirmed. The prolongation of lag period in exposed systems can be very well explained. The explanation was probably in presence of toxicants; the medium is not favorable for the growth of the exposed system might be due to the alteration of osmotic balance and second might be availability of high concentration of ions in the medium. At lower concentration of the effluent, interestingly no such lag was observed. However, at higher concentrations of the effluent prolongation of the lag phase or depletion inactive metabolism or interference of heavy metal in different biochemical metabolism leading to death of the alga was marked. Even when the alga was transferred to the toxicant free nutrient medium for recovery, it could not recover at all. This indicated the total death of the exposed alga. In this investigation, a permanent and prolonged lag can be compared with the coma stage under the influence of the toxic effluent. The growth inhibition by the effluent has been reported (Sahu, 1987) to be due to some alterations in the permeability of cell membranes, inhibition of enzyme systems, inhibition of photosynthesis and numerous other metabolic processes. We agree with the above findings. It was suggested by many authors that the effluent generally discharged into the environment should be sufficiently diluted so that the exposed systems will enjoy stimulation rather than inhibition. Stimulation of chlorophyll biosynthesis in the effluent exposed alga at sub-lethal concentrations of mercury contained effluent and inhibition of chlorophyll biosynthesis at higher doses of mercury contained effluent indicate the possible interference of mercury in biochemical processes of chlorophyll biosynthesis. While experimenting in recovery studies interesting results were obtained. Significant recovery at sub-lethal concentration of the toxicant and insignificant recovery at higher concentrations of the mercury contained toxicant during recovery period indicated dual behavior of the toxicant. Significant variations were observed in the ATPase activity of the control and effluent exposed alga. The control alga showed normal ATPase activity with the increase in exposure period. At sub-lethal / maximum allowable concentration of effluent, the exposed alga showed increase in ATPase activity compared to ATPase activity of the control alga indicating higher growth. Whereas, at higher concentration of the effluent, depletion in ATPase activity was noted in the effluent exposed algal sets. Stimulation of ATPase activity in the effluent exposed alga at sub-lethal concentrations of mercury contained effluent and inhibition in ATPase activity at higher doses of mercury contained effluent indicated the possible interference of mercury in exposed algal physiological processes of oxygen evolution and primary production and energy utilization, which was reflected as net primary productivity (NPP). Significant recovery at sub-lethal concentration of the toxicant exposed algal ATPase activity and insignificant recovery at higher concentrations of the mercury contained toxicant during recovery period indicated dual behavior of the toxicant. The regression analysis conducted on the data reported by Prusti & Panigrahi (2017a,b and 2023) clearly indicated better growth of the exposed alga in maximum allowable concentration of the effluent indicating stimulatory effect of the effluent and significant decreases in photosynthetic rate indicating inhibitory effects of the effluent. The respiration rate of the exposed alga showed dual behavior. The RR value increased in the control set with the increase in exposure period. During recovery studies, the depletion in respiration rate did not improve indicating damage to the respiratory system. The regression analysis clearly indicated better growth in maximum allowable concentration of the effluent indicating stimulatory effect of the effluent and significant decreases in respiration rate indicating inhibitory effects of the effluent. The respiration rate of the exposed alga significantly decreased and the percentage of depletion of respiration rate in the exposed alga increased with the increase in exposure period in both the selected toxicant concentrations. The respiration rate steadily decreased in effluent exposure (Prusti & Panigrahi, 2023). The effluent exposed alga was transferred to effluent free medium to test the possibility of recovery, but no recovery was noted in the GPP rate. Rather further depletion in GPP value was noted (Prusti & Panigrahi, 2022). The changes in photosynthetic rate and respiration rate in effluent exposed alga compared to control alga gave an idea of interference of mercury in the metabolic processes and attempts were made by many workers to find out the cause of the impact. We have the information that mercury along with other heavy metals are toxic and

affect the physiological processes, metabolic processes and interfere with biochemical reactions making some of the biomolecules ineffective and non functional. From the data of enzyme analysis (specifically ATPase), it was very clear that the effluent containing mercury has no significant effect on the enzyme activity at maximum allowable concentration and at LC<sub>10</sub> of the effluent. It was also observed that there was no significant difference in the enzyme activity between effluent concentrations and control at different days of exposure, as the obtained values were well within the standard deviation range. At sub-lethal concentration of the effluent maximum 6.7% increase in the enzyme activity was noted on 15<sup>th</sup> day of exposure and LC<sub>10</sub> effluent concentration 6.4% decrease on the enzyme activity was observed. From toxicological stand point the increase at lower concentration and decrease in the parameters studied at higher concentration carries a meaning and can reflect the status and impact of the toxicant. This information can be very well adopted for managing the effluent waste. The effluent can be diluted to its sub-lethal (MAC) value before discharge so that the effluent will have no effect on the organisms. But we should remember that short term exposure can provide dividends but prolonged exposure might lead to bioconcentration and metal enrichment. This heavy metal enrichment in any organism will definitely be a over burden and the heavy metal will have severe effects on the toxicant exposed organisms.

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### Conflict of Interest Statement

The authors declare that they have no conflicts of interest.

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### Reference

- Allen, M. B. and Arnon, D. I. (1955): Studies on nitrogen fixing blue-green algae, growth and nitrogen fixation by *Anabaena cylindrica*, Lemm. Pl. Physiol. Lancaster., 30, 366-372.
- APHA, (1998) (American Public Health Association): Standard methods for the examination of water and waste water. 20<sup>th</sup> Edition. APHA AWWA, WPCF. Environmental Protection Agency, USA.
- Atli, G. and Canli, M. 2007. Enzymatic responses to metal exposures in a freshwater fish *Oreochromis niloticus*. Comparative Biochemistry and Physiology, 145C: 282–287. doi: 10.1016/j.cbpc.2006.12.012.
- Belyaeva, E.A., Sokolova, T. V., Emelyanova, L.V., Zakharova, I. O (2012): Mitochondrial electron transport chain in Heavy metal-induced neurotoxicity: effects of cadmium, mercury and copper, The Scientific World J. 2012. Article ID 136063 doi: 10.1100/2012/136063.
- Bothner, M. H. and H. Carpenter (1973): Proc. 1st Ann. NSF Trace Contam. Conf., Oak Ridge Natl. Lab. 8-10 Aug. 198-210.
- Breckle, S. W., Kahle, H. (1991): Ecological geobotany / autecology and ecotoxicology. In: Behnke, H. D., ed. Progress in botany, vol. 52. Berlin / Heidelberg: Springer-Verlag, 391–406.
- Demidchik V, Sokolik A, Yurin V. (1997). The effect of Cu<sup>2+</sup> on ion transport systems of the plant cell plasmalemma. Plant Physiology 114, 1313–1325.
- Demidchik V, Sokolik A, Yurin V. (2001). Characteristics of nonspecific permeability and H<sup>+</sup>-ATPase inhibition induced in the plasma membrane of *Nitella flexilis* by excessive Cu<sup>2+</sup>. Planta 212, 583–590.
- Devi, S. R. and Prasad, M. N. V. (1999): Membrane lipid alterations in heavy metal exposed plants. In: Prasad MNV, Hagemeyer J, eds. Heavy metal stress in plants: from molecules to ecosystems. Berlin: Springer-Verlag, 99–116.
- Fimreite, N. (1970): Mercury use in Canada and their possible hazardous source of mercury contamination. Environ. Pollut., 1, 119-131.
- Fogg, G. E. (1949): Growth and heterocyst production in *Anabaena cylindrica*, Lemm. in relation to carbon and nitrogen metabolism. Ann. Bot. N.S. 13: 241-259.
- Gadd, G. M. and A. J. Griffith (1978): Microorganism and metal toxicity. Microbial Ecol., 4:303-317.
- Hartung, R. and B. D. Dinman (eds.) (1974): Environmental Mercury Contamination. Ann. Arbor. Science Publishers Inc., 349.
- Huang, X, Duan, S., Wu, Q., Yu, M., Shabala, S. (2020): Reducing cadmium accumulation in plants: structure–function relations and tissue-specific operation of transporters in the spot light. Plants (Basel) 9(2)::223.
- Janicka-Russak, M., Katarzyna. K. and Burzyn´ski, M. (2012): Different effect of cadmium and copper on H<sup>+</sup>-ATPase activity in plasma membrane vesicles from *Cucumis sativus*. Roots. Journal of

- Experimental Botany, 63 (11):4133–4142, doi:10.1093/jxb/ers097 Advance Access publication 26 March, 2012.
- Koshan, Zeyad Gattea Al-Rikabi1, Montaha Abulkareem, Al-Saffar, Adnan H. Abbas (2021): The Accumulative Effect of Heavy Metals on Liver and Kidney Functions. Medico-legal Update, January-March.Vol. 21, No. 1114.
- Kramer, H. J., Gonick, H. C. and Lu, E. (1986): *In vitro* inhibition of Na-K-ATPase by trace metals: relation to renal and cardiovascular damage,” Nephron, 44(4):329–336,
- Martineck, R. G (1970): Review of methods for determining inorganic phosphorous in biological fluids J. Am. Med. Technol., 32:337.
- Meyer, J. N., Leung, M. C. K., Rooney, J. P., Sendoel, A., Hengartner, M.O., Kisby, G. E., Bess, A. S.(2013): Mitochondria as a target of environmental toxicants. Toxicol. Sci., 134: 1-17.
- Mishra, A. K., Alaka Sahu, S. Kanwar, D. Sharma and A. K. Panigrahi (2013): Mercury pollution in and around a chlor-alkali industry: a review, The Ecoscan, Special issue-III, 349-366.
- Monteiro, S.M., Mancera, J.M., Fontainhas-Fernandes, A. and Sousa, M. (2005). Copper induced alterations of biochemical parameters in the gill and plasma of *Oreochromis niloticus*. Comparative Biochemistry of Physiology, 141C: 375–383.
- Murphy, A., Eisinger, W. R., Shaff, J. E., Kochian, L. V., Taiz, L. (1999): Early copper-induced leakage of K<sup>+</sup> from Arabidopsis seedlings is mediated by ion channels and coupled to citrate efflux. 121, 1375–1382.
- Murphy, A., Taiz, L. (1997): Correlation between potassium efflux and copper sensitivity in 10 Arabidopsis ecotypes. New Phytology 136, 211–222.
- Norrby, L. J. (1991): Why is mercury liquid? Or, why do relativistic effects not get into chemistry textbooks? J. Chem. Educ. 68, 110–113.
- O’Kelley, J. C. (1974): Organic nutrients. In: Algal physiology and Biochemistry, W. D. P. Stewart (Ed.), pp. 610. Blackwell Scientific Publications, Oxford., 610-635.
- Padhy, N. and Panigrahi, A. K. (2018a): Impact of cadmium on the toxicity and growth of a blue-green alga under experimental conditions. National Journal of Life Science, 15 (2): 133-137.
- Padhy, N. and Panigrahi, A. K. (2018b): Impact of lead on the toxicity & growth of a blue-green alga and its ecological implications. Life Science Bulletin, 15 (2): 205-208.
- Patnaik, H. (1964): Studies on nitrogen fixation by *Westiellopsis prolifica*, Janet. Ph. D. Thesis, University of London, UK.
- Prasad, P. V. D. and P. S. D. Prasad (1982): Effect of cadmium, lead and nickel on three fresh water green algae. Water, Air and Soil Pollut., 17, 263-268.
- Prusti, J. P. and Panigrahi, A. K. (2017a): Possible reclamation of solid waste of a Chlor-alkali industry by BGA and phytoremediation by plant extract under controlled conditions. National Journal of Life Science, 14 (2): 81-89.
- Prusti, J. P. and Panigrahi, A. K. (2017b): Environmental mercury discharged from a Chlor-alkali industry, its impact on biodiversity at the contaminated site at Ganjam, Odisha. Life Science Bulletin, 14 (2): 159-164.
- Prusti, J. P. and Panigrahi, A. K. (2022): Impact of Effluent of a Chlor-Alkali Industry on the photosynthetic efficiency of crop field inhabiting cyanobacteria. International Journal of Biological and Environmental Investigations, 2(1): 51-59.
- Prusti, J. P. and Panigrahi, A. K. (2023): Speciation and fate of mercury present in the solid waste and effluent of a Chlor-alkali industry discharged into the environmental segment. International Journal of Creative Research Thoughts, 11(7):111-123.
- Rath, P. (1984): Toxicological effects of pesticides on a blue-green algae, *Westiellopsis prolifica* Janet. Ph. D. thesis, Berhampur University, India.
- Rath, S. C. (1991): Toxicological effects of a mercury contained toxicant on *Anabaena cylindrica*, L. and its ecological implications. Ph. D. Thesis, Berhampur University, India.
- Sahu, A.; B. P. Shaw; A. K. Panigrahi and B. N. Misra (1986): Effect of phenyl mercuric acetate on ATPase activity of a blue-green alga, *Westiellopsis prolifica*, Janet. Microb. Lett., 33, 45-50.
- Serrano, R., Cano, A., Pestana, A.I. (1985): The plasma membrane ATPase of *Dictyostelium discoideum*. Biochimica et Biophysica Acta – Biomembranes 812, 553–560.
- Serrano, R. (1990): Plasma membrane ATPase. In: Larsson C, Moller IM, eds. The plant plasma membrane. Berlin/Heidelberg/New York:Springer-Verlag, 127–154.
- Shaw, B. P., A. Sahu, A. K. Panigrahi, (1989): Effect of the effluent from a Chlor-alkali factory on a BGA. Bull. Environ. Contam. Toxicol., 43:618-626.



- Spencer, D.F. and L. H. Nichols (1983): Free nickel ion inhibits growth of two species of green algae. *Environ. Pollut., Ser. A.*, 31:97-104.
- Sundseth, K., Pacyna, J. M., Banel, A., Pacyna, E. G., and Rautio, A. (2015): Climate change impacts on environmental and human exposure to mercury in the Arctic. *Int. J. Environ. Res. Public Health* 4, 3579–3599.
- Tangahu, B. V., Abdullah, S. R. S., Basri, H., Idris, M., Anuar, N., and Mukhlisin, M. (2011): A review on heavy metals (As, Pb, and Hg) uptake by plants through phytoremediation. *Int. J. Chem. Eng.* 2011:939161.
- Viarengo, A. (1985). Biochemical effects of trace metals. *Marine Pollution Bulletin*, 16: 153–158.
- Zhang, Y., Jaeglé, L., Thompson, L., and Streets, D. G. (2014): Six centuries of changing oceanic mercury. *Glob. Biogeochem. Cycles* 28, 1251–1261.
- Zingmark, R. C. and T. G. Miller (1975): The effects of mercury on the photosynthesis and growth of estuarine and oceanic phytoplankton. In: *Physiological Ecology of Estuarine Organisms* (Ed. F.J. Vernberg) Belle W. Baruch Library of Marine Science, University of South Carolina Press, Columbia, 3, 45-57.

