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Research Article

TOXICOLOGICAL IMPACTS OF CADMIUM CHLORIDE ON THE PRIMARY PRODUCTIVITY OF A BLUE-GREEN ALGA IN VIVO UNDER CONTROLLED CONDITIONS

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ABSTRACT

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Experiments were conducted at different concentrations of cadmium chloride to assess the impacts of cadmium chloride on the productivity of a blue-green alga, Anabaena cylindrica. The exposed alga could accumulate significant amount residual cadmium in its body within 15days of exposure and significant excretion of cadmium was observed during recovery period. Significant depletion in photosynthesis rate (PR) & respiration rate (RR) was observed in the exposed alga at higher concentrations of cadmium chloride compared to control alga. Drastic depletion in gross primary production (GPP) was computed in the experimental study with the accumulation of cadmium in the algal body. Partial recovery of the 15d exposed alga was noted after a prolonged exposure period. This crop field inhabiting tolerant alga suffers because of effluents of the paper industry which contained cadmium along with mercury and lead. The residual cadmium level increased with the increase in exposure period in all three tested concentrations. During recovery period partial recovery was noted. Prolonged recovery period indicated reversal of inhibition. The residual cadmium level decreased significantly after 30days of recovery and 60days of recovery.

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INTRODUCTION

Pulp and Paper mills are the major players to contribute heavy metals like cadmium, mercury, arsenic, copper and lead, polluting nearby water bodies. The effluent contained heavy metal mixed with river water is used for irrigation. The wastes of the effluent along with heavy metals enter in to crop fields in huge quantities. This waste affects aquatic flora & fauna. The paper mills use huge amount of fresh water drawn from water bodies in the manufacturing process and discharge significant amount of effluent as waste. Chmielowska-Bak et al., (2021) reported that "contamination of the environment with metals, their adverse impact on plant performance and transmission to the human food chain through crops and vegetables are important concerns worldwide". The paper mill effluents are discharged from the industry into the environment in and around surrounding the water bodies after simple physical and chemical treatments (Tripathy et al., 2021) indicating the need of a biological treatment. Tripathy et al., (2021) reported presence of mercury, cadmium and lead in the final discharged Paper mill effluent after physical and chemical treatment. As per technology, waste generation is a must and it is not possible to eliminate waste generation by the system (Kaur et al., 2021). But cleaner & environment friendly recycling technology can be adopted and periodically

positive modifications in the technology or alterations in the treatment technology should be planned. The solid waste and effluent from the industry are generally disposed for land filling or discharged into water bodies. Generally the toxic effluents from industry are treated before their discharge, but still they contain substantial amount of toxic substances that can cause pollution.

At present it is believed that rivers are most severely polluted by industries followed by estuaries, lakes and ocean in declining order (Sahu & Panigrahi, 2003).Cadmium was recognized many years ago to be a highly toxic element but it was not until comparatively recently that concern began to be expressed over the possible effects on crop plants, economically important plants, animals like fresh water fishes, rice crop inhabiting fishes, crabs, prawns and most important being human health after long term exposure even to low concentrations of cadmium. Mishra and Panigrahi (2023a, b) reported that cadmium chloride significantly affected the pigment content of BGA and the metal is deadly toxic. This piece of work was aimed at finding out the impact of cadmium chloride on the metabolic processes of a blue-green alga *in vivo* and its possible recovery.

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MATERIAL AND METHODS

Lethal concentration values were taken from the publication of Mishra and Panigrahi (2023a) pertaining to toxicity study: One ml of unialgal, axenic, homogenized culture was inoculated in each 150 ml flask containing 100ml of cadmium chloride solution, inside the inoculating chamber. The toxicant was diluted with sterilized nutrient medium. The homogenized algae were inoculated and the flasks were kept on culture racks. The inoculated flasks were kept inside the culture room at $28 \pm 2^{\circ}$ C and under 14 hours illumination at the intensity of 2400 ± 200 Lux and were shaken periodically daily to avoid clumping of cells. The test algae were exposed for a period of 15 days in different test media and the 15d exposed alga was allowed to recover in nutrient fresh medium without cadmium chloride.. From the toxicity testing as described by Mishra and Panigrahi (2023b), the marked X, Y and Z concentrations of the toxicants were selected for future experiments.

Anabaena cylindrica,Lemm is photo-autotrophic, unbranched, filamentous, heterocystous, blue-green alga (BGA) belongs to the family Nostocaceae. Allen and Arnon's (1955) nitrogen free medium with trace elements of Fogg (1949) as modified by Pattnaik (1964) and adopted by Sahu (1987) was most suitable basic culture for the growth of the test organisms. Oxygen evolution and carbon dioxide evolution

measurements:

The rate of oxygen evolution due to photosynthesis and oxygen consumption due to respiration was measured manometrically at 37° C and 2400 ± 200 Lux light intensity in a photo-Warburg's apparatus (New Paul, India) following the procedure of Hannan and Potouillet (1972, a), Oser (1965) and as modified by Sahu (1987). To determine the oxygen evolution rate the algal culture suspensions were centrifuged at 20^oC in 8000 rpm for 10 minutes. The supernatant was discarded and the algal mass was washed thoroughly with double distilled water and centrifuged again. The residue was suspended inside the Warburg's flask with 3 ml of substrate solution (nutrient solution and different concentration of the toxicants). The central well of the Warburg's flask containing a wick Whatman filter paper was soaked in 10% KOH solution to absorb CO_2 produced during the experiment. The experiments were performed in triplicate and the data were expressed as the mean of three observations in terms of µl of O₂ evolved / hr / algal tissue present in 100 ml culture. The carbon dioxide evolution was determined manometrically at 37[°]C in the Warburg's apparatus without supplying external light source and keeping the algal material in dark inside the Warburg's flask with 3 ml of substrate solution (nutrient solution and different concentration of toxicants). The experiment was performed in triplicate and the data were expressed as the mean of three observations in terms of µl of CO₂ evolved / hr / algal tissue present in 100 ml culture media. The obtained values were computed and tested with statistical tools to find out the validity of data and levels of significance.

RESULTS

The photosynthetic rate and respiration rate experiments were conducted at different time periods during exposure period. After 15d of exposure, the exposed algae were taken the whole content was carefully decanted and centrifuged. The residue pellet was washed 2-3 times with distilled water to remove adhered toxicants and then again centrifuged. Now the washed

pellet was taken suspended in toxicant free nutrient medium for recovery studies.

Changes in oxygen evolution rate of control and cadmium chloride exposed blue-green alga was shown in Fig.1-4, at different exposure and recovery periods. The oxygen evolution (NPP) rate increased with the increase in exposure period, showing a positive correlation ($p \ge 0.001$) in the control set. The NPP value increased from 212.5 \pm 8.8 to $356.8 \pm 14.2 \mu$ l of O₂ evolved hr⁻¹50 ml culture within 15 days of exposure. The value further increased to $432.5 \pm 13.2 \mu l$ of O_2 evolved hr⁻¹50 ml culture after 15 days of recovery. In conc.-X, the oxygen evolution rate increased at all exposure periods and recovery periods. Significant increase in the parameter was recorded after 6th day of exposure and a decrease in the rate value was recorded on 15th day of recovery, when compared to the control value. In conc.-X, the oxygen evolution rate increased from 212.5 ± 8.8 to 361.8 $\pm 8.8 \mu$ l of O₂ evolved hr⁻¹50 ml culture within 15 days of exposure, and all values were more than the control values in respective days of exposure (Fig.1).



The value further increased to $436.2 \pm 8.5 \mu l$ of O₂ evolved hr⁻ ¹50 ml culture after 15 days of recovery. In conc.-X, the oxygen evolution rate increased at all exposure periods and recovery periods and the values were more than the control values at all exposure and recovery periods. In conc.-Y, the oxygen evolution rate increased from 212.5 ± 8.8 to $311.4 \pm$ 15.6µl of O₂ evolved hr⁻¹50 ml culture within 15 days of exposure, and all values were less than the control and conc. X values in respective days of exposure (Fig.1). The value further increased to 358.4 \pm 7.4µl of O₂ evolved hr⁻¹50 ml culture after 15 days of recovery (Fig.2). In conc.-Y, the oxygen evolution rate decreased at all exposure and recovery periods and the values were less than the control and conc. X values at all exposure and recovery periods. In concentration 'Z', the oxygen evolution rate decreased from 212.5 ± 8.8 to $28.5 \pm 11.8 \mu$ l of O₂ evolved hr⁻¹50 ml culture within 15 days of exposure, and all values were significantly less than the control and conc. X and Y values in respective days of exposure (Fig.1). The value increased to $32.4 \pm 5.5 \mu l$ of O_2 evolved hr⁻¹50 ml culture after 15 days of recovery, when the exposed alga was transferred to toxicant free medium for recovery studies. In conc.-Z, the oxygen evolution rate decreased at all exposure periods and recovery periods and the values were much less than the control values at all exposure and recovery periods indicating severe damage caused to the exposed system. No recovery was noted at higher exposure periods (Fig.2). A maximum of 2.9% increase in oxygen evolution was recorded on 12^{th} day of exposure and 1.4% increase was recorded on 15^{th} day of exposure in conc-X.

Increase in oxygen evolution was marked at all exposure and recovery periods. But the increase was not significant and most of the increased values were within the standard deviation range when compared to control. In conc.-Y, the oxygen evolution rate decreased, when compared to control. A maximum of 12.7% decrease was recorded on 15th day of exposure. The oxygen evolution rate significantly decreased with the increase in exposure period and a maximum of 92% decrease was recorded on 15th day of exposure in conc.-Z. When the exposed alga of conc. Y & Z, was transferred to toxicant free medium, no recovery was marked. In conc. X, a partial recovery was recorded on 15th day of recovery (Fig.3).

The correlation coefficient analysis between days of exposure and photosynthetic rate indicated the existence of significant positive correlation in control (r = 0.991, $p \ge 0.001$); in Conc. X (r = 0.995, p \ge 0.001) and in conc. Y (r=0.992, p \ge 0.001. A negative but significant (r = -0.949, $p \ge 0.05$) correlation was marked in Conc. Z. The ANOVA test indicated the existence of non-significant difference between rows and significant difference between columns. Changes in respiration rate (CO₂ evolved) in control and cadmium chloride exposed blue-green alga were shown in Fig.3. The respiration rate increased with the increase in exposure in the control set, showing normal growth of the alga in the culture flask. The respiration rate showed the existence of a positive correlation ($p \ge 0.001$) with the exposure period. The respiration rate increased from 132.4 \pm 9.6 to 220.8 \pm 12.8µl of CO₂ evolved hr⁻¹50 ml culture within a period of 15 days. The total CO₂ evolved increased to $281.4 \pm 11.6\mu$ l of CO₂ evolved hr⁻¹50 ml culture in the recovery period after 15 days. Hence, within 30 days, the control set alga grew to such an extent that the control alga could produce $281.4 \pm 11.6\mu$ l of CO₂ evolved hr⁻¹50 ml culture (Fig.3). The respiration rate unlike photosynthetic rate showed a reverse trend. The carbon dioxide evolution increased with the increase in exposure period at conc.-X, both in exposure period and recovery period. All the exposed values were less than the control value. The carbon dioxide evolution increased from 132.4 \pm 9.6 to 191.2 \pm 9.8µl of CO₂ evolved hr⁻¹50 ml culture on 15th day of exposure (Fig.3). Significant recovery was recorded, when the exposed alga was transferred to toxicant free medium. In conc. Y, the carbon dioxide evolution showed insignificant increase from 132.4 \pm 9.6 to $158.8 \pm 11.4 \mu l$ of CO₂ evolved hr⁻¹50 ml culture on 12^{th} day of exposure and then the carbon dioxide evolution decreased to $152.2 \pm 8.5 \mu l$ of CO₂ evolved hr⁻¹50 ml culture on 15th day of exposure, showing significant depletion in carbon dioxide evolution indicating drastic effects of the toxicant (Fig.3). When the exposed alga of conc.-Y, was transferred to toxicant free medium, no recovery was marked, rather the parameter further depleted indicating acute effects of the toxicant at lower recovery periods. However, on 15th day of recovery significant recovery was noted. In case of conc.-Z, significant depletion in carbon dioxide evolution was marked when compared to control and conc. X and Y. In concentration Z, a linear decrease in carbon dioxide evolution was recorded and the percent decrease was highest on 15th day of exposure.



The carbon dioxide evolution decreased from 132.4 ± 9.6 to $18.6 \pm 8.6 \mu$ l of CO₂ evolved hr⁻¹50 ml culture on 15th day of exposure. When the exposed alga was transferred to toxicant free medium for recovery studies, no recovery was noted. Rather the carbon dioxide value further depleted to $5.4 \pm 2.6 \mu$ l of CO_2 evolved hr⁻¹50 ml culture (Fig.3). The respiration rate declined at all exposure periods and recovery periods, when compared to control values. This indicated that the toxicant is deadly toxic. At conc.-X, a maximum of 13.4% decrease in respiration rate was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 20.8% decrease was noted on 15th day of recovery. At conc.-Y, a maximum of 31.7% decrease in respiration rate was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 44.4% decrease was noted on 15th day of recovery. At conc.-Z, a maximum of 91.6% decrease in respiration rate was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 98.1% decrease was noted on 15th day of recovery. No recovery in all the three concentrations selected was noted. Rather during recovery period instead of showing any recovery, higher depletion in the respiration rate was noted. The correlation coefficient analysis between days of exposure and respiration rate indicated the existence of positive correlation in control (r = 0.986, $p \ge 0.01$); in Conc. X $(r = 0.978, p \ge 0.01)$ and in conc. Y $(r = 0.885, p \ge 0.05)$ where as a negative significant (r = -0.942, p \ge 0.01) correlation was marked in conc. Z.





The ANOVA test for the data of fig.-3 indicated the existence of non-significant difference between rows and a significant difference between columns. Changes in gross primary productivity in control and cadmium chloride exposed bluegreen alga were shown in Fig.7. The gross primary productivity increased with the increase in exposure in the control set, showing normal growth of the alga in the culture flask. The gross primary productivity showed the existence of a positive correlation ($p \ge 0.001$) with the exposure period. The gross primary productivity increased from 344.9 to 577.6µl of O_2 evolved hr⁻¹50 ml culture within a period of 15 days. The total O_2 evolved increased to 713.9 µl of O_2 evolved hr⁻¹50 ml culture in the recovery period after 15 days. Hence, within 30 days, the control set alga grew to such an extent that the control alga could produce 713.9µl of O_2 evolved hr⁻¹50 ml culture (Fig.7). The oxygen evolution increased with the increase in exposure period at concentration-X, both in exposure period and recovery period. All the exposed values were less than the control value. The gross primary productivity increased from 344.9 to 553.0µl of O₂ evolved hr ¹50 ml culture on 15th day of exposure (Fig.5). Significant recovery was recorded, when the exposed alga was transferred to toxicant free medium. In conc. Y, the gross primary productivity showed insignificant increase from 344.9 to 463.6 μ l of O₂ evolved hr⁻¹50 ml culture on 15th day of exposure, showing significant depletion in gross primary productivity indicating drastic effects of the toxicant (Fig.5). When the exposed alga of conc.-Y, was transferred to toxicant free medium, partial recovery was marked, the parameter increased indicating effects of the toxicant at lower recovery periods. However, on 15th day of recovery significant recovery was noted. In case of conc.-Z, significant depletion in gross primary productivity was marked when compared to control and conc. X and Y. In conc.-Z, a linear decrease in gross primary productivity was recorded and the percent decrease was highest on 15^{th} day of exposure. The gross primary productivity decreased from 344.9 to 47.1µl of O_2 evolved hr ¹50 ml culture on 15th day of exposure. When the exposed alga was transferred to toxicant free medium for recovery studies, no recovery was noted. Rather the gross primary productivity value further depleted to 37.8μ l of O₂ evolved hr⁻¹50 ml culture (Fig.7). The gross primary productivity declined at all exposure periods and recovery periods, when compared to control values. This indicated that the toxicant is deadly toxic. At conc.-X, a maximum of 4.3% decrease in gross primary productivity was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 7.7% decrease was noted on 15th day of recovery. At conc.-Y, a

maximum of 19.7% decrease in gross primary productivity was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 27.9% decrease was noted on 15th day of recovery. At conc.-Z, a maximum of 91.8% decrease in gross primary productivity was marked on 15th day of exposure and when the exposed alga was transferred to toxicant free medium 94.7% decrease in gross primary productivity was noted on 15th day of recovery (Fig.7A). No recovery in all the three concentrations selected was noted (Fig.7A). Rather during recovery period instead of showing any recovery, higher depletion in the gross primary productivity was noted. Fig.7 indicated the excretion of cadmium during recovery period. With the increase in recovery period more amount residual cadmium excreted and cadmium burden in the body decreased significantly which enabled chlorophyll pigment to reappear and automatically photosynthesis started. At higher exposure period and higher cadmium concentration, respiration rate almost came down to zero or an insignificant value. Significant amount of cadmium accumulated in the filaments of the alga which slowly got excreted out during recovery period (Fig.8). After 60days of recovery, significant removal of cadmium was noted and after excretion of cadmium, the exposed alga returned to normalcy and pigments reappeared. The metabolic activity of the alga was totally dependent on the cadmium burden of the exposed alga under study. The correlation coefficient analysis between days of exposure and gross primary productivity of control and cadmium chloride exposed alga, indicated the existence of strong positive correlation in control (r = 0.996, $p \ge 0.001$) and conc.- X (r = 0.990, p \ge 0.001) and conc.-Y (r = 0.992, p \ge 0.001), a negative correlation (r = -0.854, $p \ge 0.05$) in case of conc.-Z. The analysis of variance ratio test indicated the existence of a non-significant difference between rows and significant difference between columns, based on the data of figure-5. The positive results in PR value and RR value during recovery period were due to excretion of residual cadmium decreasing the metal burden on the system under study. At higher recovery period, the removal of cadmium metal from the plant body supported the survivability of the alga. After 30d exposure green dot like structures appeared and after 60days of exposure the whole white turbid mass turned green due to appearance of pigments. Appearance of pigments indicated revival of the exposed alga during recovery period.

DISCUSSION

The uptake and accumulation of cadmium by algae consists of two phases (1) adsorption of cadmium to the cellular walls and (2) penetration of cadmium into the cell. Since very little was known regarding the effects of sub-lethal concentrations of cadmium as well as mercury contained solid waste on the physiology of the freshwater blue-green alga, it was not possible to predict the detailed action on blue-green algal systems. The selectivity in mercury accumulation by plant cell might be a distinctive property of mercury including high mortality and direct uptake by the surfaces being tight bound to the acidic groups of the cell wall. A concentration and exposure time period dependent mercury uptake by blue-green algae have been observed Rath et al. (1983 a). Shaw (1987) reported effect of mercury contained industrial effluents on blue-green algae and opined that heavy metals behave drastically in a very different fashion in presence of other environmental chemicals, excluding heavy metals, and an antagonistic effect clearly prevailed with the mercury action in

presence of other chemicals. Photosynthetic pigments are known to participate in generation of energy and CO₂ fixation (Kashyap & Gupta, 1981). The chlorophylls have long been recognized as the primary light acceptors, a small portion of which acts as the primary reaction centre where light conversion occurs. Carotenoids, not only help in photosynthesis, by transferring light energy but also protect the other photosynthetic pigments, preventing photo-oxidation and providing light shielding (Krinsky, 1966). The amount of chlorophyll pigment present in the alga will be reflected on the photosynthetic efficiency of the alga. Reduction of chlorophyll content in the cadmium exposed alga probably reflected on the changes of photosynthesis rate (Mishra & Panigrahi, 2023a, b).

Algae, the most important primary producers of the aquatic environments have received least attention. Very few references are available particularly on the toxicity effects and physiological changes induced by heavy metals on algae. The review made by Whitton (1970) and Gadd & Griffiths (1978) on impact and effect of heavy metals on algae added a lot of information to the literature of algal toxicology. Algae have been shown to concentrate heavy metals to a larger extent (Trollope & Evans, 1976; and Say et al., I & II, 1977). A concentration and exposure time period dependent mercury uptake by blue-green algae have been observed by Pradhan et al (2005) and Sahu & Panigrahi (2002). Sahu (1987) reported effect of mercury contained industrial effluents on blue-green algae and opined that heavy metals behave drastically in a very different fashion in presence of other environmental chemicals, excluding heavy metals, and an antagonistic effect clearly prevailed with the mercury action in presence of other chemicals. Photosynthetic pigments are known to participate in generation of energy and CO₂ fixation (Kashyap & Gupta, 1981). The chlorophylls have long been recognized as the primary light acceptors, a small portion of which acts as the primary reaction centre where light conversion occurs. Carotenoids, not only help in photosynthesis, by transferring light energy but also protect the other photosynthetic pigments, preventing photo-oxidation and providing light shielding (Krinsky, 1966). The decrease in chlorophyll level was a result of increase in the chlorophyllase activity which gets reflected in photosynthesis rate of the exposed alga. Rath et al, 1983a,b;1985; Sahu (1987); and Sahu & Panigrahi (2002) indicated stimulation of growth, increase in pigment content, photosynthesis rate, respiration rate, and enzyme activity at lower concentrations of mercurial compounds on Westiellopsis prolifica, Janet. Cadmium & mercury at relatively low concentrations also affects the energy transfer by selectively affecting the phycocyanin in the phycobilisomes of intact cells of Spirulina, which was reported by Murty and Mohanty (1991). The photosynthetic efficiency of phycocyanin equals to that of chlorophyll-a, was reported earlier. Murty and Mohanty (1991) reported that mercury at a low concentration (3µm) caused an enhancement in the intensity of room temperature fluorescence, emitted by phycocyanin and induced a blue-shift in the emission peak of Spirulina cells indicating the alterations in the energy transfer within the phycobilisomes, whereas this phenomenon was not seen in Anacystis, in vitro. It is a common place observation that toxicity of metals showed great variations under field and laboratory conditions (Whitton, 1970). A given concentration of a metal may be more toxic to algae in the field than under laboratory conditions and vice-versa (Rai et al., 1981 b).

Hence, it becomes explicit that laboratory based information cannot solely be used to stimulate field conditions, because many environmental and nutritional factors operate to bring about metal toxicity in field conditions (Gadd & Griffiths, 1978). By measuring the photosynthetic rate, the growth can be computed and predicted indirectly as growth and photosynthesis are two intimately related terms. Eley et al. (1983) suggested that inhibition of photosynthesis might be responsible for growth retardation. Photosynthetic inhibition is mainly due to disturbances in the light energy trapping mechanism. It was observed that the inhibition of photosystem II by oxidation of cytochrome-f in the electron transport was caused by HgCl₂ in isolated chloroplast. Overnell showed that light induced oxygen evolution from the freshwater species Chlamydomonas reinhardii were very sensitive to cadmium, methyl mercury and lead. Levels of 0.8 and 1.69mg Hg l^{-1} reduced photosynthetic oxygen evolution by 50% and 90%, respectively. The inhibition of photosynthesis and respiration in plant systems by mercurial compounds was reported by Sahu et al. (1988). Gould (1975) reported that mercury influences the photosynthetic capacity of the algae by inhibiting the electron inhibitor. There were also a few reports regarding the stimulating nature of some toxicants at lower concentrations. An increase in the oxygen evolution rate at lower concentration of mercury has been reported by Rath (1984) and Sahu et al. (1988). De Filippis and Pallaghy (1976 a) hypothesized that the enhancement might be related to the lowering of the concentration of the heavy metals in the cells as they induced to divide. The author showed that mitochondrial enzyme activities and respiratory activities increased at lower concentrations of the toxicant. The activities declined significantly with the increase in the concentrations of the toxicant and the exposure period. Rath (1984) and Shaw (1987) reported the dual behavior of mercurial compounds on the growth of BGA and confirmed the dichotomous behaviour of the toxicants on living systems (Sahu, 1987 & Rath, 1991). However, this piece of work strongly agrees with the findings of the above authors. Probably the result indicated a new line of thinking, which can become a possibility in case of heterogeneous toxicants, where synergistic and antagonistic effects were expected. Here, it can be presumed that chemicals present other than cadmium probably act as a masking agent on cadmium, reducing the toxicity in turn, showing variation in the observed data. More work is essential on different live systems to confirm the synergistic and antagonistic characteristic features of the mixture toxicants. The study indicated that cadmium could affect the blue-green alga drastically causing irreparable damage. Prolonged recovery period almost 4 times more than the exposure period helped to recover the impact of cadmium. At higher recovery period, the removal of cadmium metal from the plant body supported the survivability of the alga. After 30d exposure green dot like blue-green color structures appeared and after 60days of exposure the whole white turbid mass turned blue-green due to appearance of pigments. Appearance of pigments indicated revival of the exposed alga during recovery period. With reappearance of chlorophyll pigment the photosynthetic activity initiated and the rate enhanced with time and chlorophyll pigment appearance. Cadmium should not be allowed to escape and be available in the environment for protection of flora, fauna, human beings and the environment.

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Conflict of Interest Statement

The authors declare that they have no conflicts of interest.

Author Contribution Statement

Prof. A.K. Panigrahi: Conceptualization, planning and execution of the project, field visit, original draft preparation, supervision, reviewing and editing. Research work conducted by Sri Saroj K. Mishra analysis and related experimental work. Mishra contributed reagents, glassware, field related work, calculation and finalization of data.

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