

## Biosorption efficacy of living and non-living algal cells of *Microcystis aeruginosa* to toxic metals

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### Abstract

The existence of metallic elements in the aquatic environment is recognized to cause acute destruction to aqueous life. This study depicts the prospective application of cyanobacterial strains of *Microcystis aeruginosa* as a sorption material of toxic elements, aluminium (Al), and cadmium (Cd) from aqueous solutions. Algal samples were revealed to the metal solution, a noticeable modification change in cell wall structure surface occurred. The Fourier-transform infrared (FTIR) analysis illustrated the reality of carboxyl, carbonyl, and hydroxyl moieties, which are liable for the uptake of essential and nonessential elements aluminium and cadmium, respectively. The results showed the ability of *Microcystis aeruginosa* to uptake Al and Cd at the optimal temperature, light, and pH by living and non-living cells in the concentration of 20 ppm and stimulated antioxidant resistance against oxidative stress. This finding divulged that *Microcystis* could be utilized as an efficient bio-sorbent for the elimination of these ions, especially Cd from freshwater.

**Keywords:** antioxidant enzymes; bioremediation; microcystis; X-ray

### Introduction

The algal cell has the aptitude to bioaccumulation supreme essential and nonessential metallic elements even though the range of bio-accumulation contrasts reckoning on the bio-availability of the metallic elements, the algal cell into deliberation, also concentrations to that they're visible (Francis *et al.*, 1985; Roane *et al.*, 2015; Narendrula-Kotha *et al.*, 2019). The activity of humans has significantly affected the dissemination of metallic elements in the natural involving air, water, and sediment. Several heavy elements are vital for life, like copper (Cu), zinc (Zn), aluminium (Al), and nickel (Ni) since they are important catalysts for enzyme metabolism (Han *et al.*, 2020).

In disparity, some elements are not only nonessential elements for life but are also have lethal effects as cadmium (Cd), and lead (Pb). Overall, nonessential, and poisonous heavy elements are not cracked by aquatic microorganisms (Han *et al.*, 2020). To abolition of metal ions from an aqueous solution, copious techniques

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are applied. These incorporate exchange of ions, ion precipitation, extraction of plant, and electro-dialysis (Yu *et al.*, 1999; Yadav *et al.*, 2019).

Heavy metals are lethal effectiveness and lead to the contamination of water and prospective problems in public health (Islam *et al.*, 2008). Consequently, it is critical to eradicating heavy elements like Pb, Al, and Cd that are frequent heavy elements obtained in wastewater discharge. Cd is an organically stirring element; it can observe in element ores, also like organic sediments in the earth. High altitudes of Cd were observed in the field of shellfish and other filter-feeding organisms. Excessive quantities of Cd ravaged the kidney in addition to that activate stones of the kidney to form and osteoporosis to ensue (Organization, 2003).

In rejoinder to mounting public anxieties on multiplied incidence and enormosity of elements contamination, plentiful procedures have been established to evaluate the hazards, including biological functions and individual health, to revelation industrial toxic discharges (Al Ketife *et al.*, 2020). Nevertheless, these techniques are either useless or costly in eradicating concentrations of trace elements (Yadav *et al.*, 2019).

Various permutations of environmental factors can instigate the deficiency of oxygen and poisonousness in freshwater. Plants will be polluted, and some aquatic animals inept to stay alive except for algae. Consequently, the capability of the wastewater medication system to endure and eradicate poisonous is of substantial consequence (Utomo *et al.*, 2016).

Biosorption that utilizes the biological techniques is a comparatively new application for eradicating trace concentrations of heavy elements from contaminated water. Bio sorbent substances like naturally occurring seaweeds are generally rarer pricey than fundamental technologies (Holan *et al.*, 1993). Microalgae have substantiated to acquire high elements binding abilities owing to the existence of polysaccharides, proteins, or lipids on the algal cell wall surfaces. Numerous other microbial sources have been used by various investigators (Itoh *et al.*, 1975; Niu *et al.*, 1993; Chang *et al.*, 1997).

*Microcystis* cyanobacteria are predominantly valuable because of their widespread dispersion. One of the major anxieties relate to *Microcystis* sp. bloom is its capability to generate the toxin microcystin (MC) (Babica *et al.*, 2006; Mohan *et al.*, 2020). *Microcystis* sp. has a strong capability to adjust to the environment.

The vital characteristics of biosorption and bioaccumulation of metals whereas the convention of living micro-organisms is frequently efficacious in dealing with poisonous organic pollutants, living micro-organisms in conformist biological treatment systems have been infrequently convenient in the treatment of solutions that have heavy metal ions. After the metal ion concentration converts to too high or adequate elements are absorbed by the microorganism, the micro-organism's metabolism is interrupted, hence instigating the micro-organism to die. This disorder does not exist if non-living micro-organisms (biomass) originated from microorganisms are applied to adsorb element ions from solution/effluents (Michalak *et al.*, 2013). *Microcystis* is a unique group of concern here, due to their tolerance and interaction with element ions (Rzymiski *et al.*, 2014a).

Most microalgae possess a negative charge on their surface. The main mechanism of metallic cation sequestration includes the creation of complexes among element ions and electronically rich functional groups on the surface and within the pores of algae. The carboxyl groups of alginates play a major role in forming complex structures (Rivas *et al.*, 2009). The cells of *M. aeruginosa* keep a highly developed capacity to acclimate to undesirable stress as increasing in the size of the cell wall and the growth rate of the *Microcystis* strain restricted. Moreover, the enzymatic defence system and the oxygen radicals play an important role in enzymatic and detoxifying reactions (Vichnevetskaia and Roy, 1999).

The survey validates that *Microcystis* sp. endures high concentrations of heavy metals; it similarly has a great capability to remove elements from aquatic environments and can be valuable for bio-remediation of these elements from ecosystems (Çabuk *et al.*, 2005; Mohan *et al.*, 2020). The absorption process of the cells (living or non-living cells) is diverse (Ilhan *et al.*, 2004). The use of dead biomass eradicates the dilemma of toxicity from dissolved metals and remove the problem of nutrient supply. Though, algal living cells may exhibition a broader range of mechanisms for metal accumulation, such as extra-transport and extracellular complex formation.

This survey intended to evaluate the toxic effects of Cd(II) and Al(III) on the algal cell *M. aeruginosa* by measuring the antioxidant enzymes and to evaluate its bioaccumulation capacity as a function of the concentration of the metals under the different condition and the ability of living and non-living cell to contact with the metals.

## Materials and Methods

### *Micro-alga isolate*

*Microcystis aeruginosa* was obtained from the National Research Centre, Cairo, Egypt. Algal culture incubated at optimal growth conditions (pH 7.0, Temp. 25 °C, the irradiance of 60  $\mu\text{mol m}^2/\text{s}$ ) delivered by white, fluorescent lamps 14:10 h light: dark photoperiod for 1-2 weeks which is suitable for photosynthesis. Al and Cd stock solution was added to UV-sterilized BG-11 media at pH 7, Temp. of 25 °C. The cultures of *M. aeruginosa* were centrifuged at 3000 rpm for 15 min, after that, the cells were washed with filtered H<sub>2</sub>O. The washed cells of *M. aeruginosa* were dehydrated at 60 °C. The dehydrated biomass is hoarded in a sealed bottle.

### *Experimental design*

Cadmium nitrate Cd (NO<sub>3</sub>)<sub>2</sub>•4H<sub>2</sub>O and aluminium nitrate Al (NO<sub>3</sub>)<sub>3</sub> were used as a source of metals in this work, all chemicals were used as received without any further purification and were obtained from Sigma-Aldrich. The cyanobacterial cells were centrifuged (10 minutes, 3.000 rpm) and dissolved in filtered sterilized H<sub>2</sub>O in 250 mL conical flasks. The initial cell density was 8.5×10<sup>6</sup> cells mL<sup>-1</sup>. Metals were added in concentrations of 5, 10, 15 and 20 ppm of Cd or/and Al.

*M. aeruginosa* cells were preserved with Cd and Al under optimal growth conditions at different periods of contact time (3 h, 6 h, 24 h, and 48 h). This analysis was intended to know the most effective treatment times of the process of metal biosorption. These time intervals with the effect of different acidity (pH 5, 6, 7, and 8) on metal sorption and the efficacy of the biosorption process on the equivalent metal concentrations (5, 10, 15, or 20 ppm). Every experiment was carried out in triplicate.

### *Characterization of biomass*

#### *Fourier Transform Infrared (FTIR) analysis*

FTIR spectrophotometer JASCO 70 type made in Japan was used to examine the dry algal biomass of *M. aeruginosa* before and after the bioaccumulation process. This process was performed to identify the functional groups on the strained surface.

#### *Scanning electron microscopy (SEM) and energy dispersive x-ray (EDX) spectroscopy*

2.5% glutaraldehyde was employed to fix algal cultures for 4 h. and rinsed with sterile distilled water 4 times. As a final point, metal stubs mounted all samples and coated with gold. An Inspects' Model scanning electron microscope (JEOL JSM-5500 LV SEM) was utilized to show the images, and energy dispersive X-rays, coupled to SEM was used (Dwaish *et al.*, 2011).

### *Enzyme activities estimation*

Enzyme extract was prepared as termed by (Mukherjee and Choudhuri, 1983). Peroxidase (POD) activity was evaluated by the guaiacol oxidation process (Li *et al.*, 2013). Superoxide dismutase (SOD) activity was ascertained by the tetrazolium reduction method (Li *et al.*, 2013). UV absorption method was used to determined catalase (CAT) activity (Li *et al.*, 2013).

#### *Estimation of lipid peroxidation*

Lipid peroxidation was assessed by measuring the quantities of malondialdehyde (MDA) created by the thiobarbituric acid reactive substance (TBARS) as illustrated by Li *et al.* (2013), utilizing 0.5% (w/v) thiobarbituric acid (TBA) in 20% (w/v) trichloroacetic acid (TCA). The absorbance was measured at 532 and 600 nm by using the extinction coefficient of  $155 \text{ mM}^{-1} \text{ cm}^{-1}$ .

#### *Study the elect of elements by using living and non-living algal biomass*

The biosorption ratio was assessed as follows: Biosorption (%) =  $[(C_i - C_f)/C_i] \times 100$ , where  $C_i$  - "initial concentration", and  $C_f$  - "final concentration". The non-living algal cells attained by the heat killing method by placing the algal cultures on a water bath at  $50^\circ\text{C}$  for 2h, then centrifuged to get a pellet, which then put in 50 ml culture media at 20 ppm (Inthorn *et al.*, 2002).

#### *Statistical analysis*

One-way ANOVA was applied to detect significant differences and the standard deviation (SD). among different concentrations of heavy metal. Differences among individual means were revealed by Tukey's post hoc multiple range test  $p < 0.05$ .

## **Results and Discussion**

#### *Functional groups in cyanobacteria (Microcystis aeruginosa)*

FTIR analysis was validated as an efficient method for the identification of both essential and nonessential metals bio-sorption on cyanobacteria (*Microcystis aeruginosa*). The spectrum of free algae and algae loaded with cadmium and aluminium metals demonstrated the variety of functional groups observed on the cell wall surface of analysed bio-sorbents algal cell (Table 1 and Figure 1).

Figure 1 shows a checked wave number from  $4000 \text{ cm}^{-1}$  to  $400 \text{ cm}^{-1}$ , before and after exposition to a 10 mL solution containing Al and Cd at concentrations of 20 ppm. The FTIR examination publicized which functional groups will be capable of the adsorption of the toxic metals.

Figure 1 illustrates the control test with the groups related to the hydroxyl O-H group, lignin, and acetyl group. The groups at  $3400 \text{ cm}^{-1}$  and  $2900 \text{ cm}^{-1}$  are doled out to O-H-N, H stretching, and C-H stretching, individually, and it exists in control and loading samples. After adsorption of cadmium and aluminium, slight shifts were noticed in the absorption peak frequencies between  $400$  and  $4000 \text{ cm}^{-1}$ . Where these peaks ( $3427.85$ ,  $2867.02$ ,  $1044.26$ ,  $839.85 \text{ cm}^{-1}$ ) are shifted to lower frequencies ( $3421.02$ ,  $2857.02$ ,  $1037.52$ , and  $830.45 \text{ cm}^{-1}$ , respectively) after loading the algae with Cd (II) and Al (III). This was perchance owing to the coordination of the heavy metals to an active functional group causing a vibration frequency below the selected range. This is attributed to the attachment of the heavy metals to an active functional group causing a lower vibration frequency. The peaks detected attest to the existence of carboxylic acids, hydroxyl ions, and carbonyl groups, with others on the algal surface, as suggested in the literature (Davis *et al.*, 2003; Gupta *et al.*, 2006; Rangel *et al.*, 2016).

Additionally, the focal functional groups that can biosorption are the hydroxyl, carbonyl, carboxyl, sulfonate, amide, imidazole, phosphonate, and phosphodiester functional groups as declared by (Volesky, 2007). Several of these functional groups appear in the strain of *M. aeruginosa* and possess the decent capacity to interact with toxic elements. Furthermore, the binding of the element with biopolymers primarily materialized in the peptidoglycan and layer of the cell surface (Lin *et al.*, 2005; Mohan *et al.*, 2020).

**Table 1.** Fourier-transformation infrared peaks and their possible assignments of *Microcystis aeruginosa*

| Peak position before loading (cm <sup>-1</sup> ) | Peak position after loading Cd (ii) | Peak position after loading Al (iii) | Possible assignment                                 |
|--|-------------------------------------|--------------------------------------|---|
| 3427.85  | 3421.02                             | 3421.02                              | O-H str, alcohols/ phenols, N-H, amines             |
| --   | 3286.11                             | --                                   |   |
| 2925.48  | 2925.48                             | 2925.48                              | C-H str, carboxylic acids                           |
| 2867.02  | 2857.02                             | 2857.02                              |   |
| 1654.62  | 1657.52                             | 1657.52                              | C=N, C=O vib, C=C, N-H bend                         |
| 1550.49  | 1543.74                             | 1546.65                              |   |
| 1450.21  | --                                  | 1450.21                              |   |
| 1387.53  | 1390.39                             | --                                   | O-H bend, S=O str, CH <sub>3</sub> bend and ss, N-O |
| --   | 1269.90                             | 1269.90                              |   |
| 1044.26  | 1037.52                             | 1033.66                              | C-N str, aliphatic amines                           |
| 839.85   | 830.45                              | 839.85                               |   |
| --   | 624.82                              | 621.93                               | -C≡C-H: C-H bend alkynes, C-Br str, alkyl halides   |

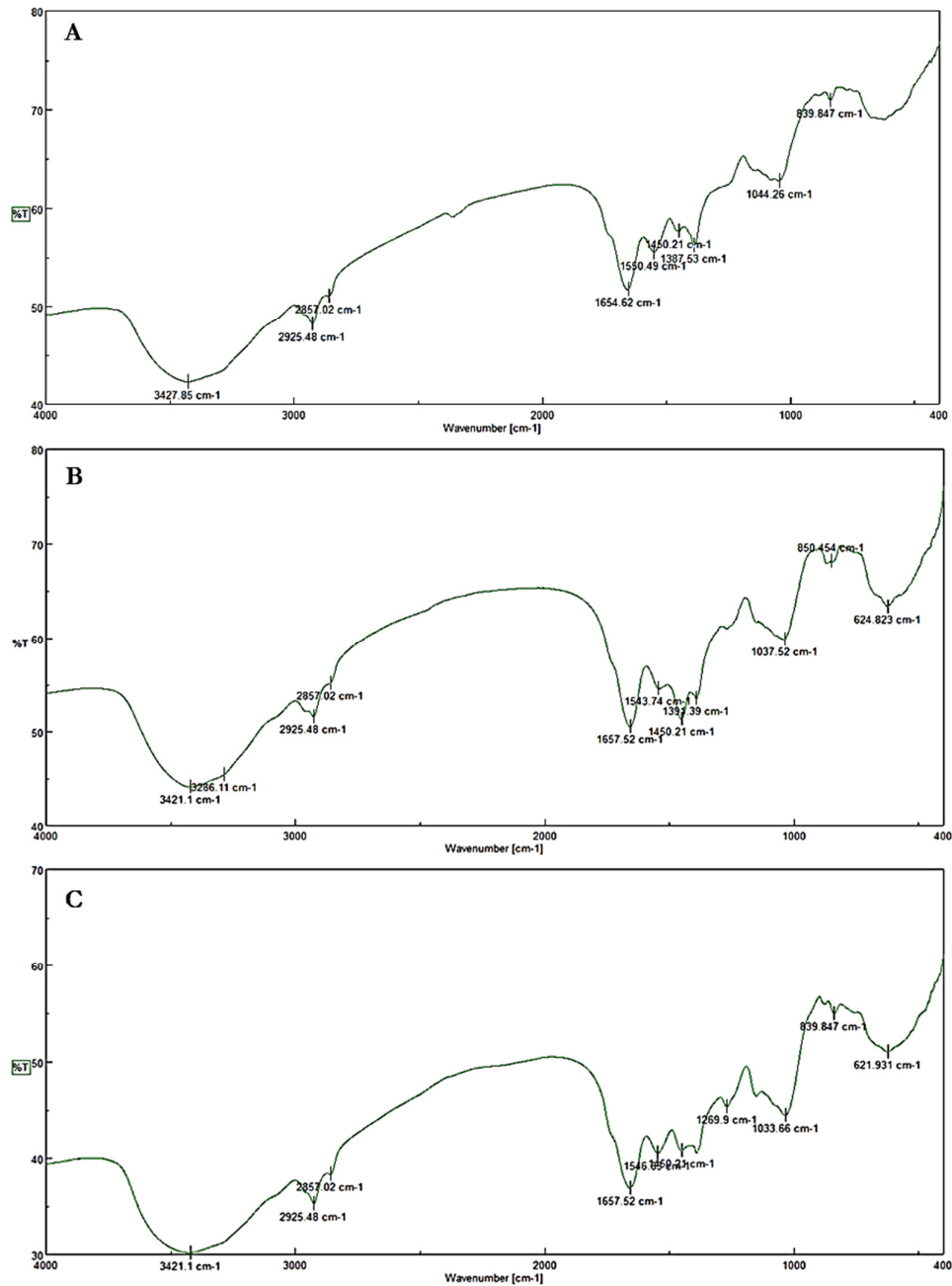
Additionally, the focal functional groups that can biosorption are the hydroxyl, carbonyl, carboxyl, sulfonate, amide, imidazole, phosphonate, and phosphodiester functional groups as declared by Volesky (2007). Several of these functional groups appear in *M. aeruginosa* and possess the decent capacity to interact with toxic elements. Furthermore, the binding of the element with biopolymers primarily materialized in the peptidoglycan and layer of the cell surface (Lin *et al.*, 2005; Mohan *et al.*, 2020).

To study the effect of elements on fresh alga *M. aeruginosa* we used SEMT and EDX to determine the capacity of this microorganism to sorption heavy metal (Al and Cd). In this autopsy, SEMT can be utilized to envision the morphological surface of *M. aeruginosa* before and after metal binding, permitting for direct inspection of any variations that exist (Figure 2). To distinguish the chemical composition and variations concerning the pure and metal-loaded algal surface in the *M. aeruginosa* we used both SEM and EDX.

The pure sample of *M. aeruginosa* indicates corroborate of smooth structures arranged in quite a regular pattern on the surface, whereas these are not existing in the Al and Cd-loaded images.

Insightful morphological alterations were observed in the structure of the surface wall (Figure 2). The cell wall structure has fluctuated in diameter, and they got to be enlarged and destroyed with a twisted surface. Related alterations were detected in other algae (Narayani *et al.*, 2016). It was obvious that Cd and Al have the ability to biosorption on the active sites of *M. aeruginosa* cells.

The majority of the algae were able to absorb heavy metals either by biosorption or bioaccumulation. There are several studies and reviews on the sorption of metal ions into marine algae (Dwaish *et al.*, 2011) and species of freshwater algae with various efficiencies of removal. This heavy metal uptake ability may be due to the functional groups found on the cell wall surface that can serve as binding sites.



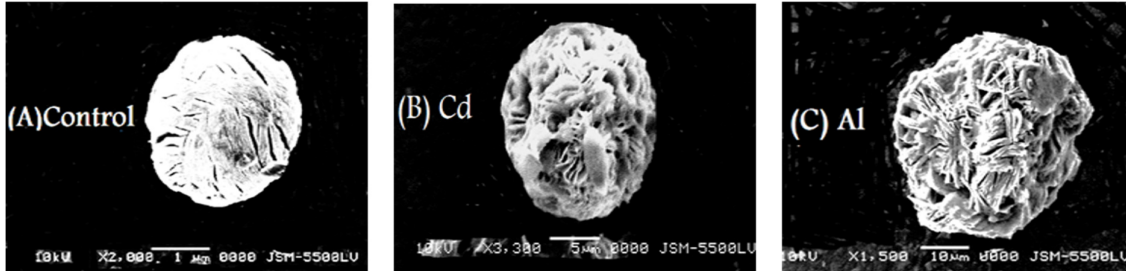
**Figure 1.** Fourier-transformation infrared of control (A), cadmium (B) and aluminium (C) loaded alga *Microcystis aeruginosa*

*Impression of SEM/EDX results*

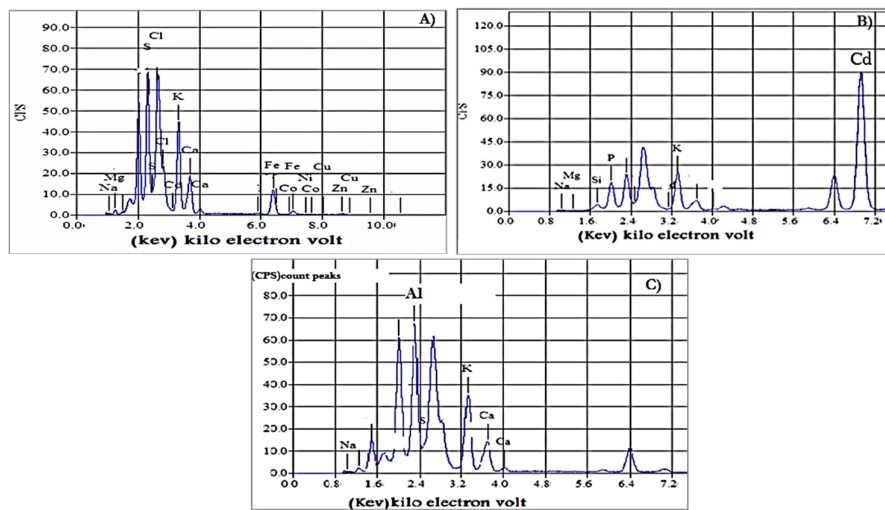
While samples were exposed to metal solutions, the ion-exchange mechanism may occur by replacement (Figures 3 and 4). Cd has been stated to have a primary impact on Ca homeostasis as it can displace Ca from binding sites and further indicates a strong interference with the movements of K<sup>+</sup> and Ca<sup>+2</sup> in the cells (Di Toppi and Gabbrielli, 1999).

Singh *et al.* (2013) anticipated that the replacement of these ions with metal cations distorted the nature of the cross-linking anticipated to stronger electrostatic and coordinative bonding among the metal and the negatively charged groups in the cell wall polymers.

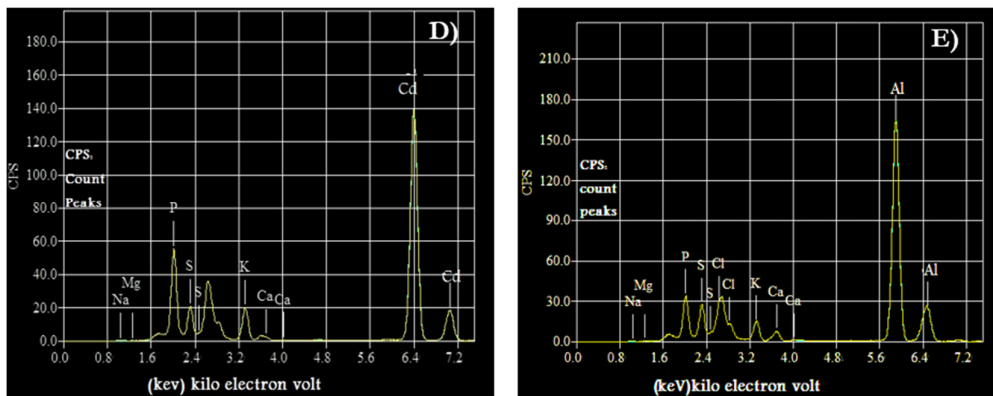
The Al and Cd loaded elements spectra of *M. aeruginosa* revealed the changing of alkali and alkaline earth metal peaks (K and Ca) (Raize *et al.*, 2004). This is validated by data in this survey, wherever the replacement of calcium, magnesium, and sodium and potassium peaks were similarly detected, as illustrated in Table 2. Accordingly, it seems that alterations in the cross-linking behaviour associated with the metal's coordination sphere caused obvious morphological variations in the surface of the algal cell.



**Figure 2.** Scan electron micrographs of pure *Microcystis aeruginosa* control (A), after Cd (II) uptake (B), and after Al (III) uptake (C).



**Figure 3.** Energy- dispersive X-ray spectroscopy of Control (A), Cd (II) (B) and Al (III) (C) -loaded by living cells of *Microcystis aeruginosa*



**Figure 4.** Energy-dispersive X-ray spectroscopy of Cd (II) (D) and Al (III) (E) -loaded by non-living cells of *Microcystis aeruginosa*

**Table 2.** Elemental peaks (Cd (II) & Al (III)) in the energy dispersive X-ray spectroscopy of *Microcystis aeruginosa*

|                               | Pure sample | Cd (II) loaded | Al (III) loaded |
|-------------------------------|-------------|----------------|-----------------|
| <i>Microcystis aeruginosa</i> |             |                |                 |
| Sodium (Na)                   | Na          | *              | *               |
| Magnesium (Mg)                | Mg          | *              | *               |
| Potassium (K)                 | K           | K              | *               |
| Calcium (Ca)                  | Ca          | Ca             | Ca              |
| Cadmium (Cd)                  | *           | Cd             | *               |
| Aluminium (Al)                | *           | *              | Al              |

\* Band not observed. (Raize *et al.*, 2004).

Oxygen radicals are generated in case of environmental stresses. These oxygen radicals ought to be rummaged for the preservation of normal growth. A lot of plant systems demonstrating that environmental stresses change the quantities and the activities of enzymes immersed in scavenging oxygen radicals (Chang *et al.*, 1997). The activity of CAT, SOD, and POD and the content of MDA in *M. aeruginosa* strain utilized in our analysis increased when the algal cell exposure to the toxic metals, especially in the case of Cd metals. The results showed that antioxidant enzymes might play vital parts in eradicating the undue reactive oxygen species (ROS).

Cd is known to induce enhanced ROS production in general (Pandey *et al.*, 2009; Gutiérrez-Martínez *et al.*, 2020) as illustrated in Table 3. A strong antioxidative defence and strong redox homeostasis have been associated with metal tolerance (Sharma and Dietz, 2009) and antioxidant accumulation in stressed algal cells (Noctor *et al.*, 2012; Gutiérrez-Martínez *et al.*, 2020).

**Table 3.** Effect of essential and non-essential elements on the activity of catalase (CAT), superoxide dismutase (SOD), peroxidase (POD) (unit  $\text{min}^{-1} \text{g}^{-1}$  fresh weight), and the content of malondialdehyde (MDA) (nmol  $\text{g}^{-1}$  fresh weight) of *Microcystis aeruginosa* cultures

| Treatment                          | (ppm) | CAT                     | SOD                     | POD                     | MDA                      |
|------------------------------------|-------|-------------------------|-------------------------|-------------------------|--------------------------|
| Control                            | 0     | 10.00±1.00 <sup>a</sup> | 5.00±0.20 <sup>a</sup>  | 2.67±0.42 <sup>a</sup>  | 157.00±6.08 <sup>c</sup> |
| Cd (NO <sub>3</sub> ) <sub>2</sub> | 20    | 13.00±0.50 <sup>b</sup> | 6.03±0.17 <sup>c</sup>  | 3.67±0.47 <sup>ab</sup> | 201.10±1.68 <sup>d</sup> |
| Al (NO <sub>3</sub> ) <sub>3</sub> | 20    | 10.50±0.70 <sup>a</sup> | 5.44±0.10 <sup>ab</sup> | 2.77±0.49 <sup>a</sup>  | 170.00±10 <sup>c</sup>   |

<sup>abc</sup> and <sup>d</sup> mean in the same row within different letters, different significantly (at  $P \leq 0.05$ ).

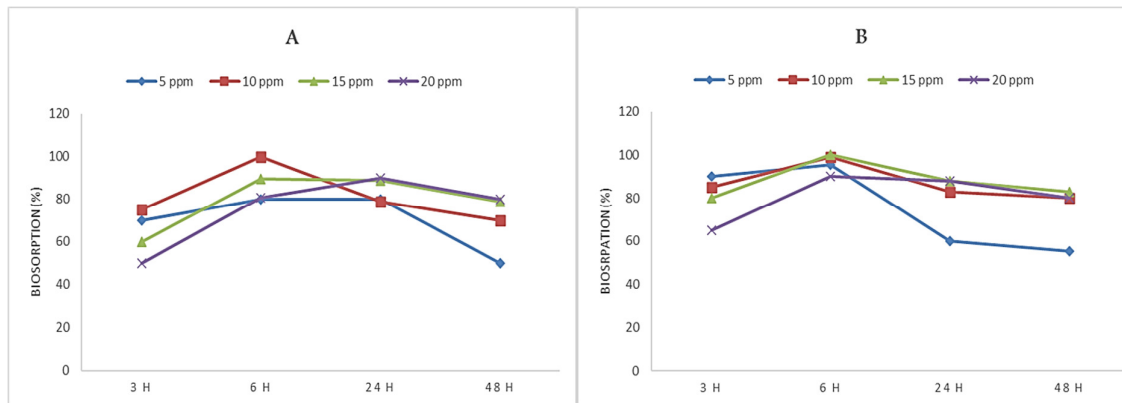
All values were expressed as Mean ±SD, n = 3.

#### *Biosorption of Al & Cd at optimal growth conditions*

*M. aeruginosa* strain proved a definite capacity to take each, Al and Cd at optimal conditions (pH 7.0, temp. 25 °C, the irradiance of 60  $\mu\text{mol photons m/s}$ ), leading to a considerable decreasing in element concentrations. The potency of element sorption relied on the time of cultivation and the initial concentration of Al or Cd though the sorption rate of the tested sample exceeded 50% (Figure 1). Al illustrated that, the highest effective reduction (reaching 100%) in the concentration of metal ions after 6 h of exposure to the toxic metal. Total sorption of Al was observed in samples with an initial metal content of 10 ppm, whilst 15 ppm and 20 ppm, sorption capacities attained 89.4% and 80.5%, respectively. A shorter time of incubation (3 h) was valuable only for the lowest Al concentration. Longer incubation (1 day and 2 days) considerably modified the sorption potency (Figure 5).

Based on these outcomes, we tend to assume that 6 h of dealing with Al was the most actual time. Contrasted to Al, removal of Cd at the same growth conditions was typically slower after 6h of contact. The percentage of removal in 5 ppm, 10 ppm, 15 ppm, and 20 ppm treatments resulted in 95.4%, 99.2%, 100.0%, and 90.2% biosorption of metal, respectively. Together with shorter (3 h) and longer time (24h and 48h) Cd contact illustrated less efficient biosorption process (Figure 5). Our outcomes implicitly demonstrated that tolerance of *M. aeruginosa* to Al and Cd could be based on metal sorption (Rzymiski *et al.*, 2014b) and

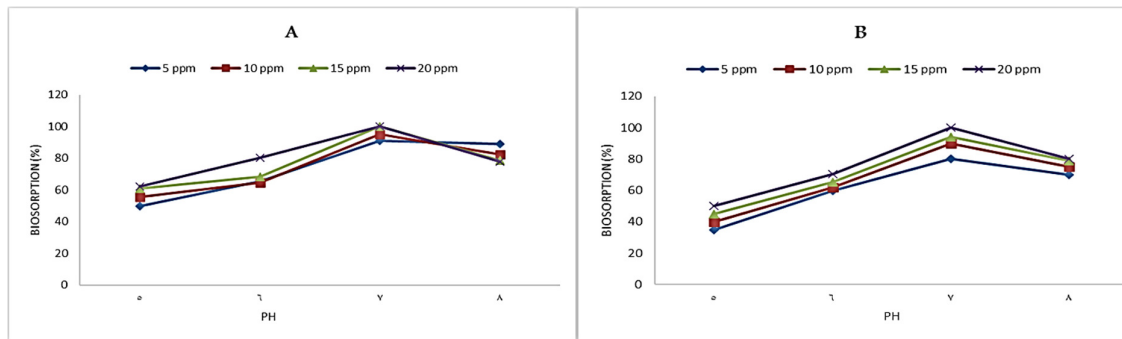
subsequent contact time. After three hours of treatment, uptake of metals is likely to be a dominant process. As found in alternative studies, Al uptake in cyanobacteria also as alternative algae, occurs more rapidly than Cd (Romera *et al.*, 2007). The difference in the ionic size of metals, ionic radii, electrode potential, and affinity to the functional groups on the bio-sorbents illustrated the capacity of the biosorption process (Sekhar *et al.*, 2004). Typically, in our analysis, by increasing the initial metal concentrations, the biosorption capability increase, specifically for Cd. This is due to the reality that an increase in metal content provides a larger driving force to overcome all mass transfer resistance between solid and aqueous phase, thus resulting in higher metal ion adsorption (Edris *et al.*, 2014). In our survey, *M. aeruginosa* strain displayed a greater potency of metal biosorption.



**Figure 5.** The effect of treatment time on biosorption of Al (A) and Cd (B) by *Microcystis aeruginosa*

#### *Biosorption of Al & Cd at different pH values*

pH of the solution considers a vital factor for restricting the biosorption process. *M. aeruginosa* strain has distinct groups of amino, hydroxyl, carboxyl, and sulphate on the cell wall, which are affected by variations in the pH values (Ibrahim *et al.*, 2016). The efficacy of biomaterials to sorption processes was evaluated at distinct levels of acidity as illustrated in Figure 6. Ascending and descending pH can significantly change the biosorption of studied elements. The lowest biosorption rate occurred at low pH (Figure 6). From our results, biosorption of Al was less influenced by pH. The sorption capacities increased at optimal pH at different algal concentration. It decreased by a mean of 65% at pH 5 and 40% at pH 6 ( $p < 0.01$ ) under the lowest concentration of algal cell ( $5 \text{ mg L}^{-1}$ ). At pH 8 biosorption of Al was in turn minimized by 30% ( $p < 0.05$ ). The capability of Cd to the biosorption process was more susceptible to pH alterations (Figure 6). At lower acidity and lower algal concentration, the capacity of *M. aeruginosa* to metal removal decreased significantly. Alkaline pH additionally reduced the efficiency of Cd sorption by approximately 25.0% at pH-8 ( $p < 0.05$ ). Our data prove that metal sorption by *M. aeruginosa* strain can be shifted to the lowest values at acidic pH. This observation attributed to the proposed mechanism of microalgae prevention to low extracellular pH by reducing metals cations uptake and simultaneously accelerated anions uptake (Rai *et al.*, 1996; Krüger *et al.*, 2012). Also, this diminution in metal remediation at low pH was observed in previous studies of cyanobacteria (Kumar and Gaur, 2011). It is worth knowing that acidity can be raised the toxicity of metals to microalgae due to the prevalence of the free metal ion (Fathi and Al-Omair, 2006). To increase the efficiency of bioaccumulation and biosorption, the potential use of *M. aeruginosa* should therefore be preceded by an adjustment of pH.



**Figure 6.** The effect of pH on biosorption of Al (A) and Cd (B) by *Microcystis aeruginosa*

*Bioaccumulation of metals at optimal growth conditions and different algal cell state (Living and non-living algal cells)*

*M. aeruginosa* cell exhibited a distinctive talent to acceptance each of Al and Cd at optimal conditions (pH=7.0 and Temp=25 °C), subsequent a remarkable decline of metal conc. in aqueous solution. The proficiency of metal sorption varied according to the time of incubation and the initial concentration of Cd (II) or Al (III). The assumptions indicated that (Cd<sup>++</sup>), and (Al<sup>+++</sup>) percentage uptake was 69.50% and 60.77% in living cell *M. aeruginosa*. These percentages were significantly increased to 98.44% for Cd (II) and 92.40% for Al (III) in non-living cells of *M. aeruginosa* (Table 4). There was an association concerning the natural habitat of algae and their assets of the uptake of the heavy metal that agrees with the results of (Rabsch and Elbrächter, 1980; Ahmad *et al.*, 2020).

**Table 4.** Percentage of essential and non-essential elements uptake by living and non-living cells of *Microcystis aeruginosa* cultures

| Treatment                         | Metal conc. (ppm) | Metal uptake by living cell % | Metal uptake by non-living cell % |
|-----------------------------------|-------------------|-------------------------------|-----------------------------------|
| Essential elements<br>Al (III)    | 20                | 60.77                         | 92.40                             |
| Non-essential elements<br>Cd (II) | 20                | 69.50                         | 98.44                             |

Lately, the awareness has altered to non-living algal cells for the removal of heavy metals. In distinction to live cells, we found that the metal sorption capability of dead cells may be more. Higher desirability of non-living cells for metal ions contrasted to living biomass that might probably be owed to deficiency of struggling protons generated throughout metabolism.

## Conclusions

In this finding, bio-sorption and bio-monitoring study give substantial information concerning the appropriateness of cyanobacteria *M. aeruginosa* as a bio-sorbent, and a bio-monitor for the nominated heavy metals in the solution, the optimal temperature, light, and pH for this process are supplied. From this scrutinize, the algae are also germane as a bio-indicator because it can accumulate metals to a satisfactory degree. The metal uptake is augmented by using a non-living cell. Further studies are required to extend the bio-sorption abilities of biomass and develop applicable biological technologies in wastewater therapy. The *Microcystis aeruginosa* that is isolated from the polluted region can pick up cadmium and aluminium. The uptake of those metals is depended on the metal concentration and their physiological standing. The great

variations in the responses of the alga to the heavy metals may recommend the metal concentration in the water- samples in their habitat.

### Authors' Contributions

Conceptualization: EAA; Methodology: EAA; Validation: EAA; Formal analysis: EAA; Investigation: EAA; Data curation: EAA; Funding acquisition: AAHAL; Writing: EAA; Review and editing: AAHAL, ME; All authors read and approved the final manuscript.

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### Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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