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# Bioremediation of Chromium (III) from Tannery Effluent using Microalgae

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**Abstract:** Heavy metal (HM) pollution has slowly but surely crept into the list of direst issues being faced by mankind. It usually results in a myriad of complications for the environment as well as human health. Conventional techniques for the treatment of HM pollution are sometimes not effective under certain conditions. Treatment of HM pollution is currently being explored by utilizing biological activity of plants, algae, fungi, etc. Positive outcomes have been demonstrated, especially by algae. Thus, phycoremediation has arisen as an alternative, green method of solving this issue. Green algae were isolated from Kolavai lake and then cultivated in Erlenmeyer flasks under room conditions. After growth, the cells were harvested by centrifugation and filtration and then dried for 24 hr at high temperatures. Powdered biomass was then added to aqueous solutions containing Chromium at different concentrations with a fixed dosage. Results showed that 30ppm was favored for $Cr^{3+}$ uptake by biosorbent. Next, the effect of pH was studied on adsorption rate. Results indicated that as the pHincreased, the $Cr^{3+}$ uptake was found to have decreased, thus confirming low pH favors $Cr^{3+}$ sorption. AAS results had indicated that biosorbent was effective in $Cr^{3+}$ even at low concentrations and thus provided a possible alternative to conventional techniques.

**Keywords:** Algae, Bioremediation, Wastewater, Heavy Metals, Chromium, Biosorption

## 1. Introduction

Due to the negative consequences it is producing all across the world, Heavy metals (HM) pollution is an issue that is only getting worse and is now a major worry. Through a variety of human activities, including agriculture, the metals industry, inappropriate waste management, fertilizers, and pesticides, HMs are released into the surroundings [1]. Moreover, HMs are

produced naturally by erosion, infiltration, and volcanic activity [2]. When these contaminants are released into the environment, they build up in the soil, water, and atmosphere. Diseases affecting the gastrointestinal system, the lungs, the heart, the kidneys, the hemopoietic system, and the nervous system can all be brought on by HM exposure.

Both anthropogenic (man-made) and geogenic (natural) sources can release HMs [3]. Volcanic eruptions, rock weathering, etc. are some of the natural sources that release HMs [4]. The burning of fossil fuels, car exhaust, mining, agriculture, the incineration of solid and liquid waste, and atmospheric pollution are all examples of anthropogenic sources of HMs [2, 5]. Both natural and manmade sources of HMs can build up in the environment, and plants can absorb these metals through tainted soil and water. HM water contamination is caused by a variety of human activities, including the use of fossil fuels, car exhaust, mining, agricultural practices, and waste incineration. Moreover, HMs are produced naturally by volcanoes, thermal spring activity, erosion, infiltration, and other processes [2]. Human health can suffer greatly as direct consequence of HM pollution. The kidneys, liver, skin, and cardiovascular system are only a few of the human body's organs where HMs can be toxic [6]. Metals that are toxic to humans can enter the body through the skin, digestive system, or lungs. It has been demonstrated that toxic metals can harm DNA and membranes, as well as interfere with protein synthesis and enzyme activity [7]. Mercury, Lead, Chromium, Cadmium, and Arsenic are the HMs that are most frequently linked to health issues [8]. The harm that these HMs can do to membranes and DNA, the disruption of protein and enzyme activity, and the development of gastrointestinal, respiratory, cardiovascular, reproductive, renal, hemopoietic, and neurological illnesses are all possible effects of exposure to them [2]. Developmental delays, brain impairment, and kidney disease have all been related to mercury exposure. Anemia, hypertension, and brain and nervous system damage can all result from lead exposure. Exposure to chromium can result in liver and kidney damage, stomach cancer, lung cancer, and other types of cancer. Prostate cancer, kidney damage, and lung damage can all be brought on by cadmium exposure. Skin sores, skin cancer, liver, kidney, and lung damage can all result from exposure to arsenic [8].

Algae can be bioengineered using methods that are particular to plants, and the majority of algal biotechnological research and applications will focus on creating novel products or algae that can synthesis commercial products at a competitive price. In addition, engineered microalgal biochar for sustainable remediation of the emerging pollutants from wastewater [9]. As they have a relatively straightforward genetic structure and are significant economically, the form of algae known as cyanobacteria has been the subject of most advancement in algal genetic engineering [10]. Algal-based membrane reactor for the remediation of emerging contaminants from wastewater: mechanism, synthesis and technological advancement also elaborated [11]. Many lucrative items could be produced using algal biotechnology [12]. Polypeptides, polysaccharides, organic acids, biofuels, and other products with added value can all be made from algae. Cultivation of microalgae at the cheap cost by utilizing wastewater as nutritional source for the production of algal biomass [13]. Over 200 species of algae are utilised globally in various

industries as food, feed, and fertilizers. The removal of environmental pollutants from water, soil, etc. using living organisms like bacteria, fungi, microalgae, and plants is a process known as bioremediation. A sustainable method of managing environmental contamination is bioremediation. It can be used in addition to physicochemical methods to completely solve the problems posed by environmental contaminants. Pesticides, solvents, explosives, oil and other petroleum products, and others are among the toxins that bioremediation can remove [14].

## 2. Materials and Methods

### 2.1 Sample Collection

An unknown sample of green algae species was collected from Kolavai lake, South Chennai (Latitude- 12.721060, Longitude- 79.982920). The algae was then washed with distilled water and inoculated in agar plates in order to isolate pure single colonies.

### 2.2 Algae Culture

BG-11 medium, also known as blue green algae 11 culture medium, was prepared by appropriately weighing the 11 reagents specified above, mixing them with distilled water followed by autoclave at 121°C for 15 min.

Table.1. Contents of BG-11 growth medium and their required concentration

| REAGENT | QUANTITY (mg/L) |
|---|---|
| Sodium Nitrate (NaNO3) | 1500.0 |
| Magnesium Sulphate (MgSO4.7H2O) | 75.0 |
| Ferric Ammonium Citrate | 12.0 |
| Boric Acid (H3BO3) | 2.861 |
| Dibasic Potassium Phosphate (K2HPO4) | 39.0 |
| Cupric Sulphate (CuSO4.5H2O) | 0.07 |
| Sodium Carbonate (Na2CO3.H2O) | 20.0 |
| Calcium Chloride (CaCl2.2H2O) | 27.0 |
| Sodium Molybdate (Na2MoO4.2H2O) | 0.391 |
| Manganese Chloride (MnCl2.4H2O) | 1.81 |
| Zinc Sulphate (ZnSO4.7H2O) | 0.222 |
| EDTA | 1.0 |
| Trace Metals | 1mL/L |

The algae sample collected was then inoculated on a petri dish containing the BG-11 agar medium. The plates were allowed to incubate for two weeks and were regularly monitored in order to obtain pure colonies. When appropriate growth was achieved, pure colonies were observed under the microscope. The colonies were then inoculated in conical flasks containing

the BG-11 growth medium. The flasks were kept in an area that received sufficient light at approximately 26°C. In order to obtain sufficient amount of biomass, the culture medium was scaled up to a 15 L vessel after a month.

## 2.3 Biomass Extraction and Drying

After appropriate amount of biomass was obtained, the extraction process was initiated by centrifuging the medium for a period of 10 min at 4500 revolutions/min. The supernatant was then discarded while the pellets were collected in a petri plate. The collected wet biomass was then kept in a hot air oven at a temperature of 60°C for a time period of 24 hr for drying. Using a mortar and pestle, dried biomass was grounded and then sieved through a 75 mesh size sieve in order to obtain uniform powdered biomass.

## 2.4 Synthetic Wastewater Preparation

Industrial grade Chromium sulphate was obtained from Ejaz Tanning Company, Ltd. located in Ambur. To half a litre of distilled water, 188.5 mg of Chromium sulphate was added in order to obtain 100 ppm aqueous chromium stock solution. From the stock solution, various solutions of differing concentrations were prepared for the experiment.

## 2.5 Adsorption Study

Chromium aqueous solutions of varying concentrations (15, 30 and 45 ppm) were prepared by diluting the stock solution. Previous studies had found the optimum adsorbent dosage to be 1g/L [15]. Five conical flasks were taken to which 100 mg of powdered biomass was added to four of these flasks. The following solutions were added to each flask for 100 mL; - distilled water, 15ppm, 30 ppm, 45 ppm and 100 ppm Chromium aqueous solution. The flasks were kept in an orbital shaker maintained at 130 rpm for 2 hours. Next, in order to separate the biomass from the solution after adsorption, the solutions were subjected to centrifugation at 4500 rpm for 10 min followed by filtration through a Wattman filter paper. The filtrate was then analysed by UV- visible spectroscopy at 400-580 nm. Once the optimum concentration had been elucidated, a second round of adsorption studies was conducted, this time by varying pH as a parameter. Five flasks containing synthetic wastewater had their pH adjusted from 4 – 8 by adding 0.1 N HCl or 1.0 M NaOH. 100 mg of powdered biomass was added to all the flasks. The flasks were kept in an orbital shaker maintained at 130 rpm for 2 hours. The solutions were then centrifuged at 6000 rpm in order to allow the fine particles to settle down thus, separating the supernatant.

## 3. Results and Discussion

Initially, 2 L culture vessel was theorized to yield 1g dry weight of algae. However, after drying, the weight recovered was a measly 0.13g. Thus, there was a need to scale up the culture vessel to a 15 L tank, which resulted in a total wet weight of 50g obtained after harvesting algae. After drying, the dry weight recovered was found to be 1.6g, thus crossing the 1g threshold required for the experiment. Analysis of the supernatant by UV- visible spectroscopy determined the extent to which chromium remained in the solution. The following data was obtained from subjecting the solutions to UV- visible spectroscopy before and after adding the dry biomass (Figure 1a, b)

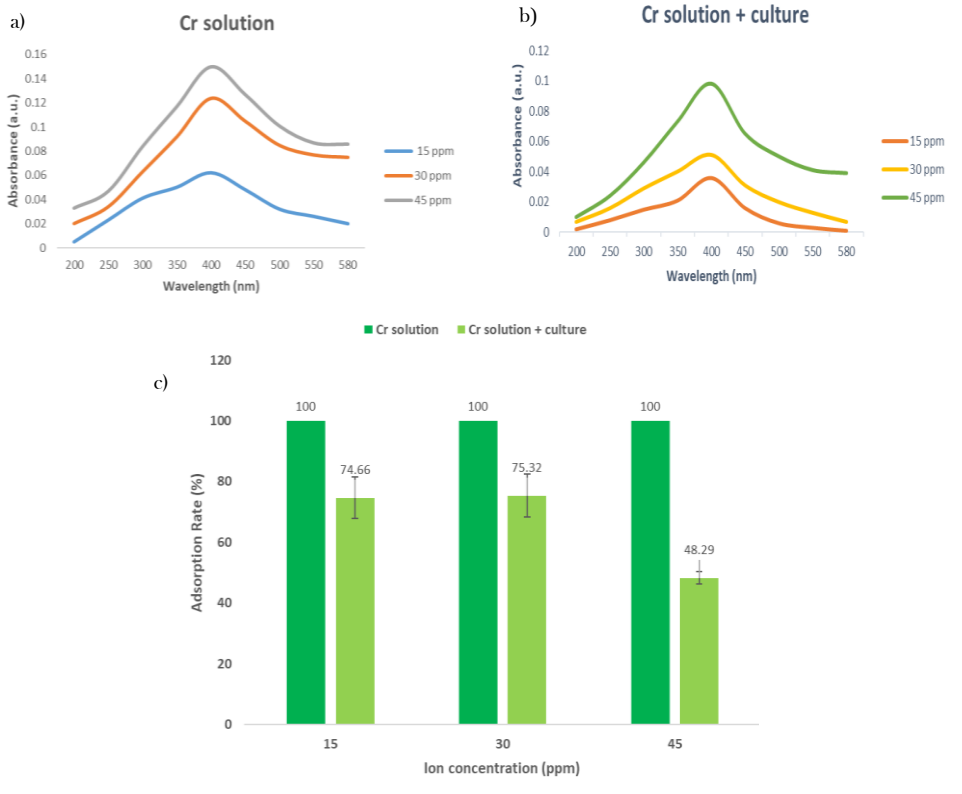


**Figure 1.** Graph showcasing synthetic wastewater (a); after addition of culture (b); Comparison between graphs of Cr solution and Cr solution + culture for each ion concentration (15, 30, 45) (c).

The data obtained from the table was then used to compare graphs of a particular ionic concentration in order to make a prediction about the adsorption rate. From the line graphs, a bar graph was generated that determines the adsorption efficiency of each particular ion concentration (Figure 1c). From the graph, 30 ppm ion concentration was determined to be the optimum concentration for Cr uptake by biosorbent with the dosage 1g/L. While both 15 ppm

and 30 ppm showed high adsorption rate for biosorbent, there was a significant decrease at 45ppm. This was attributed to the fact that 1g/L of adsorbent dosage will have a fixed number of binding sites that can take up $Cr^{3+}$ from the solution. Initially, an increase in initial concentration was accompanied by an increase in removal rate by the biosorbent. However, any further increase in initial concentration did not result in an increase in adsorption rate. This was attributed to the fact that the fixed number of binding sites becomes saturated and are not able to adsorb any more ions from the solution [16]. 30 ppm was selected for the next round of experiments to be conducted in order observe the influence of pH. The parameter was adjusted using a pH meter to values 4, 5, 6, 7 and 8. After centrifugation, the following data was obtained from subjecting the solutions to UV- visible spectroscopy before and after adding the dry biomass. The data was then used to generate a line graph in order to visualize the results (Figure 2 a,b)

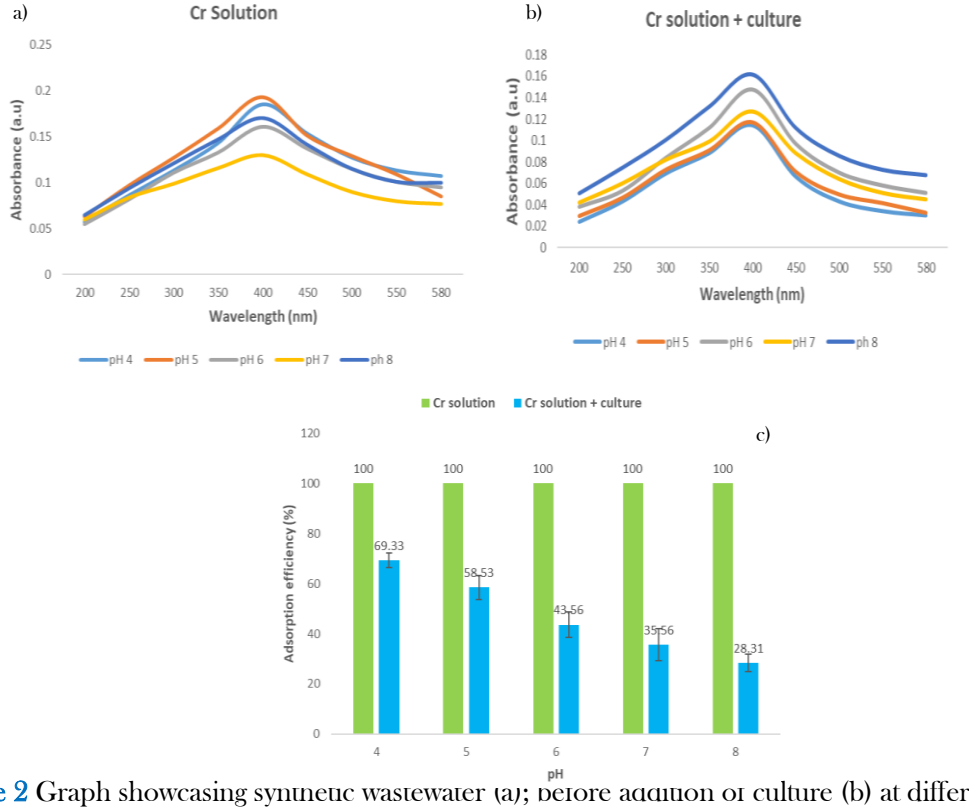


**Figure 2** Graph showcasing synthetic wastewater (a); before addition of culture (b) at different pH (4, 5, 6, 7 & 8); Comparison between graphs of Cr. solution and Cr solution + culture for each particular pH (4, 5, 6, 7, 8) at each pH with standard deviation The data obtained from the table was then used to compare graphs of a particular pH in order to make a prediction about the adsorption efficiency. From the line graphs, a bar graph was generated that determines the adsorption rate of each particular pH (Fig. 2c).

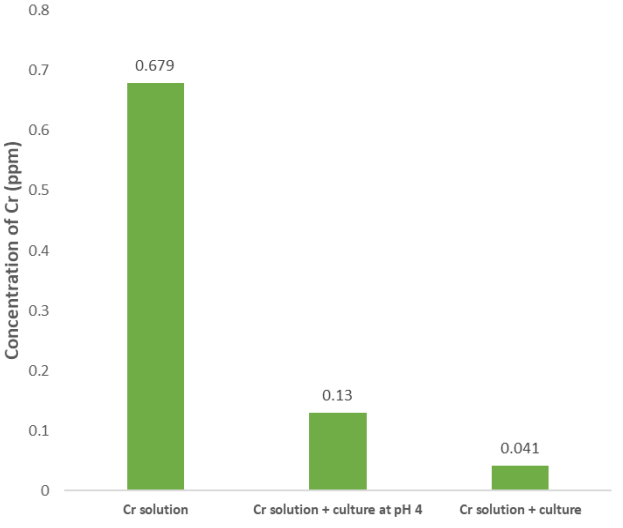


**Figure 3.** Graph showcasing Cr adsorption at the optimized conditions

Results showed that as the pH increased, this was accompanied by a decrease in adsorption rate. This could be attributed to the fact that an increase in pH facilitates the precipitation of metal ions [16]. According to Kumar *et al.*, (2011), Cr ions exist as $HCrO^{4-}$ or $Cr_2O_4^{2-}$ ions at low pH which interface with the binding sites of the adsorbent. As the pH keeps rising, conversion of Cr ions to $Cr_2O_4^{2-}$ is favoured [17]. Experiments carried out in Nithya *et al.*, (2019) presented that the high uptake of Cr ions at low pH is due to the presence of $HCrO^{4-}$ ions which predominates other forms between pH levels 1 to 4 [18]. The binding sites get deprotonated as the pH rises, which causes them to repel negatively charged chromate ions such as $Cr_2O_4^{2-}$ which predominate at higher pH levels. While the effect of pH of the present work was conducted between 4 to 8, Nithya *et al.*, (2019) subsequently found out the optimum pH for Cr biosorption to be pH 2, which consequently, was the pH at which $HCrO^{4-}$concentration remained maximum. Atomic Adsorption Spectroscopy (Thermofisher Scientific, USA) [18]. Three samples were tested using AAS; standard $Cr^{3+}$ solution, $Cr^{3+}$ solution with the addition of biosorbent, and $Cr^{3+}$ solution with the addition of biosorbent maintained at pH 4. The results were then compiled to indicate the amount of $Cr^{3+}$ remaining in each sample using a calibration curve constructed using standard solutions of 1.25, 2.5, and 5ppm.Thus, it is clear from the data that maximum amount of HM removal occurred at the sample to which biosorbent was added and where the pH was not adjusted (i.e. pH < 2.5). While adsorption rate is still high, adjusting the sample to pH 4 was not as effective at $Cr^{3+}$ uptake as the sample maintained in its own natural pH (Figure 3).

## 4. Conclusion

Synthetic wastewater of varying concentrationswas prepared using industrial grade chromium sulphate. Dried culture was added to synthetic wastewater which was kept in an orbital shaker at 130 rpm for 2 hr. Biomass was separated from solution and then weighed for comparison with biomass pre-adsorption. UV- Visible spectroscopy was conducted on the samples to determine the concentration at which adsorption rate was maximum.Dried culture was again added to synthetic wastewater of varying pH which were kept in an orbital shaker at 130 rpm for 2 hr. UV- Visible spectroscopy was conducted on the samples to determine the pH at which adsorption rate was maximum.AAS was then conducted to find out the exact amount of $Cr^{3+}$ remaining in the sample. It was soon established that $Cr^{3+}$ uptake remained high when pH remained low due to existence of $HCrO^{4-}$ ions that have a high affinity towards biosorbent. It was seen that biosorbent could remove $Cr^{3+}$ even at low concentrations, thus overcoming the issue faced by some conventional techniques.

## References

[1] J. Briffa, E. Sinagra, R. Blundell, Heavy metal pollution in the environment and their toxicological effects on humans. Heliyon, 6(9), (2020) e04691. https://doi.org/10.1016/j.heliyon.2020.e04691

[2] M.L. Sall, A.K.D. Diaw, D. Gningue-Sall, S. Efremova Aaron, J.J. Aaron, Toxic heavy metals: impact on the environment and human health, and treatment with conducting organic polymers, a review. Environmental Science and Pollution Research, 27, (2020) 29927-29942. https://doi.org/10.1007/s11356-020-09354-3

[3] V. Masindi, P. Mkhonza, M. Tekere, (2021) Sources of Heavy Metals Pollution. Environmental Chemistry for a Sustainable World, Springer, Cham. https://doi.org/10.1007/978-3-030-80334-6_17

[4] M. Pujari, D. Kapoor, 1-Heavy metals in the ecosystem: Sources and their effects. Heavy Metals in the Environment, (2021) 1-7. https://doi.org/10.1016/B978-0-12-821656-9.00001-8

[5] A.S. Mohammed, A. Kapri, R. Goel, (2011) Heavy Metal Pollution: Source, Impact, and Remedies. Biomanagement of Metal-Contaminated Soils. Environmental Pollution, 20. https://doi.org/10.1007/978-94-007-1914-9_1

[6] S. Mitra, A.J. Chakraborty, A.M. Tareq, T.B. Emran, F. Nainu, A. Khusro, A.M. Idris, M.U. Khandaker, H. Osman, F.A. Alhumaydhi, Jesus Simal-Gandara, Impact of heavy metals on the environment and human health: Novel therapeutic insights to counter the toxicity. Journal of King Saud University-Science, 34(3), 101865. https://doi.org/10.1016/j.jksus.2022.101865

[7] D. Witkowska, J. Słowik, K. Chilicka, Heavy Metals and Human Health: Possible Exposure Pathways and the Competition for Protein Binding Sites. Molecules, 26(19), (2021) 6060. https://doi.org/10.3390/molecules26196060

[8] N. Rama Jyothi (2021) Heavy Metal Sources and Their Effects on Human Health. Heavy Metals -Their Environmental Impacts and Mitigation. https://doi.org/10.5772/intechopen.95370

[9] G.G. Satpati, D. Kundu, R.C. Rajak, S. Gupta, J.W. Kim, M. Davoodbasha, Algal-based membrane reactor for the remediation of emerging contaminants from wastewater: Mechanism, synthesis and technological advancement. Algal Research, 79, (2024)103465. https://doi.org/10.1016/j.algal.2024.103465

[10] I. Rasul, A. Farrukh, H.S. Muhammad, M. Saima, R. Azhar, M. Anam, A. Muhammad, A.A. Muhammad, N. Habibullah, Chapter 8 - Algae Biotechnology: A Green Light for Engineered Algae, Editor(s): Khalid Mahmood Zia, Mohammad Zuber, Muhammad Ali. Algae Based Polymers, Blends, and Composites, (2017) 301-334. https://doi.org/10.1016/B978-0-12-812360-7.00008-2

[11] G.G. Satpati, A. Devi, D. Kundu, P.K. Dikshit, S. Saravanabhupathy, R. Banerjee, R.C. Rajak, M.R. Kamli, S.Y. Lee, J.W. Kim, M. Davoodbasha, (2024). Synthesis, delineation and technological advancements of algae biochar for sustainable remediation of the emerging pollutants from wastewater-a review. Environmental Research, 258, 119408. https://doi.org/10.1016/j.envres.2024.119408

[12] M. Fabris, R.M. Abbriano, M. Pernice, D.L. Sutherland, A.S. Commault, C.C. Hall, L. Labeeuw, J.I. McCauley, U. Kuzhiuparambil, P. Ray, T. Kahlke, P.J. Ralph, Emerging Technologies in Algal Biotechnology: Toward the Establishment of a Sustainable, Algae-Based Bioeconomy. Frontiers in Plant Science, 17, (2020) 279. https://doi.org/10.3389/fpls.2020.00279

[13] R. Sami-ullah, D. MubarakAli, S.B. Hisham, A. Hesham, S. Usman, A.T. Aqeel, A.S. Aliyu, W. Al-Alaya, M.S. Azim, (2023) Utilization of Wastewater as Nutritional Source for the Production of Algal Biomass. International Journal of Energy Research, (2023) 8. https://doi.org/10.1155/2023/9939252

[14] P. Singh, V.K. Singh, R. Singh, A. Borthakur, S. Madhav, A. Ahamad, A. Kumar, D. Bahadur Pal, D. Tiwary, P.K. Mishra, Chapter 1 - Bioremediation: a sustainable approach for management of environmental contaminants, Abatement of Environmental Pollutants, (2020) 1-23. https://doi.org/10.1016/B978-0-12-818095-2.00001-1

[15] M.E. Goher, A.M. Abd El-Monem, A.M. Abdel-Satar, M.H. Ali, A.E. Hussian, A. Napiórkowska-Krzebietke, Biosorption of some toxic metals from aqueous solution using non-living algal cells of Chlorella vulgaris. Journal of Elementology, 21(3), (2016) 703–714.

[16] G. Sibi, Biosorption of chromium from electroplating and galvanizing industrial effluents under extreme conditions using Chlorella vulgaris. Green Energy & Environment, 1(2), (2016) 172-177. https://doi.org/10.1016/j.gee.2016.08.002

[17] A. Kumar, D. Suman, B. Chiranjib, D. Siddhartha, Adsorptive removal of chromium (VI) from aqueous solution over powdered activated carbon: Optimisation through response surface methodology. Chemical Engineering Journal, 173(1), (2011) 135-143. https://doi.org/10.1016/j.cej.2011.07.049

[18] K. Nithya, A. Sathish, K. Pradeep, S.K. Baalaji, Algal biomass waste residues of Spirulina platensis for chromium adsorption and modeling studies. Journal of Environmental Chemical Engineering, 7(5), (2019) 103273. https://doi.org/10.1016/j.jece.2019.103273

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