



Bacteria Mediated Biosorption of Hexavalent Chromium - A Review

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ABSTRACT

A good living standard is always a major priority of every human being in this world. The continuous industrialization, urbanization, and an increase in population has directly or indirectly accounted for the endless mass pollution of various basic needs such as water, air and land. This has created a significant havoc in the global environment leading to several health issues both for humans as well as other life forms. Hence, there is an imperative need for environment friendly procedures to help remove heavy metals from the ecosystem. The biosorption process has been considered as a supremely promising and appealing approach for heavy metal elimination from industrial wastewater. Recent research has focused on using biosorption to remove hexavalent chromium using various biosorbents. The major advantage of such a biological process includes good efficiency in nature and application of low cost biosorbents which can be reused. The present chapter provides an overview of the removal of hexavalent chromium using different types of bacteria extracted from various environments.

Keywords: Biosorption, Biosorbents, Biomass, Hexavalent chromium, Pollution, Wastewater

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INTRODUCTION

Heavy metal wastes generated from industry are the principal source of human-induced contaminants in the environment. Trace metals like lead, mercury, cadmium, and chromium also have a substantial effect on health. The occurrence of such wastes in the air, water or soil even in trivial amounts can threaten the long-term health of all living beings [1]. Trace metals namely Pb (lead), Hg (mercury), Cd (cadmium) and Cr (chromium) are of substantial consideration due to their ability to build up through the tropic levels of the ecosystem inducing a harmful ecological impact [2].

The massive industrial utilization of the non-biodegradable heavy metal Chromium (VI) is a serious environmental concern as it can accumulate in living organisms. This makes it imperative to treat such wastes prior to releasing them into the atmosphere. In order to serve this purpose efficiently and cost-effectively, the biosorption method can be employed [3]. Biosorption is identified as a process accomplished with the help of active/inactive microbial biomass called biosorbents that bind and concentrate the heavy metals. This approach when used for removal of Cr (VI) is prompt, reversible, passive and promising in contrast to the other conventional methods used like chemical precipitation, ion exchange, reverse osmosis etc.. The cell wall components of the microorganisms assist them in carrying out the biosorption process by adsorbing the inorganic contaminants. Major contribution to biosorption is attributable to the anionic ligand groups such as carbonyl, hydroxyl, phosphoryl and sulfhydryl [4].

HEXAVALENT CHROMIUM

Heavy metal contamination is most commonly associated with copper (Cu), cadmium (Cd), lead (Pb), nickel (Ni), zinc (Zn) and chromium (Cr). Amongst these Chromium (Cr) is of major concern to life. Natural sources of it include soil, plants, animals, rocks, gases and dust particles. It subsists in a range of oxidative states with valency from +2 to +6, of which 0, +3 and +6 are accounted to be the most stable states. Trivalent and hexavalent forms of lead are becoming increasingly concentrated in the environment

due to human activities. Chromium contaminants in ground water are present in the form of oxyacids and oxyanions of hexavalent chromium, which is very soluble, toxic and mobile in its oxidation state. Hexavalent chromium can be carcinogenic depending on its chemical form, valency and solubility under certain conditions [5].

Hexavalent chromium intake is possible by all cell types wherein it is reduced to the trivalent form inside the cells. This trivalent chromium is capable of directly binding to the DNA besides formation of DNA-protein cross linkages resulting in mutagenesis. Furthermore, hexavalent chromium ion reduction within the cells drives the synthesis of unstable compounds like Cr (V) and Cr (IV) and thiol radicals. Upon reaction with oxygen, hexavalent chromium forms reactive oxygen species (ROS), for instance, hydroxyl or hydrogen peroxide radicals. All these conditions, finally direct towards DNA damage and gene mutations [6].

The limitation for maximum authorized discharge levels into surface and portable water set by regulatory authorities is below 0.05 mg. Exposing humans to Cr (VI) are linked to respiratory cancers. Man-made contamination of fresh water resources occurs when industrial effluents are released into them without appropriate treatment. Chromium occupies a prominent position among industrial alloys due to its use in a wide array of processes, including plating, textile manufacture, petroleum refining, leather tanning, wood preservation, and pulp processing. In addition to steel and metal alloys, cement, galvanized plastics, tanneries, paints, fertilizers, and fungicides, this metal is also used in the fabrication of these products. According to research, Cr (VI) is approximately one hundred times more toxic than Cr (III), a crucial component of human metabolism for maintaining glucose, cholesterol, and triglyceride levels. Chromium (III) is very much capable to oxidize into Cr (VI), the more toxic form [7].

The hydrological cycle is responsible for a number of environmental crises, including water pollution. Manufacturing industries release toxic chemicals and contaminants from different sources at diverse levels. Chromate VI (Hexavalent) is one of the most toxic heavy metals and is proven to be toxic at levels as low as parts per billion (ppb). Its properties are so advantageous that it stays for prolonged periods in various physical and chemical constituents of the ecosystem. The species of chromium generated by each anthropogenic source are contrasting [8].

Chemical precipitation and coagulation with lime are some of the conventional methods used to remove hexavalent chromium from industrial effluents. These techniques accompany some drawbacks including greater energy requirements, insufficient removal of metals and heavy load of toxic sludge wastes attributed to the chain of reagents used in the treatment processes. As a result, seeking a suitable substitute solution and developing techniques that help in the exclusion of these contaminants is a necessity. The application of biosorbents in coupled or uncoupled form is currently being used to eliminate Cr (VI) [4].

BIOSORBENTS

A biosorbent used must be predominantly aim at immobilizing the metal ions. Therefore, the selection of a pertinent biosorbent is considered as the primary phase in formulating biosorbents. The elements governing the selection criteria include rigidity, ability to endure intense operating conditions, cost of material and treatment, provision and steady supply. In other words, the process designed must be cost-effective and the origin of the biomass used for the metal biosorption must be scrutinized. In certain cases where the biomasses are cultivated to exploit their biosorption ability, the expense and unpredictability in retaining a constant supply of biomass must be taken into account. Even the waste generated from certain industries like food, fermentation and pharmaceuticals can be utilized to treat the metal wastes using the potent microbial species of *Saccharomyces*, *Corynebacterium* and *Streptomyces* [9].

The high surface to volume ratio and electronegative charges on microbial cell walls result in brilliant nucleation sites. It is believed that all research activities start with native microorganisms derived from areas polluted by heavy metals, hence it is very important screen and isolate potential bacteria with unique metabolic competencies [10].

The mechanism governing the biosorption process relies on several other intrinsic and extrinsic conditions. For treatment of heavy metals, choosing a suitable biosorbent is the foremost requirement, followed by other features like viability of the cell, metabolic products, specific growth velocity, nutrient/substrate needs, and growth environment involving temperature, pH and dissolved oxygen. The type of the effluent compounds and their configuration should be investigated, as well as the type and concentration of metallic species. There is evidence that a large number of microbes display a biphasic response, when presented with low concentrations of inhibitory compounds, they grow and when they are presented with high concentrations of inhibitory compounds, they shrink. This corresponds to the lowest concentration of the metal contaminant which may lead to the suppression of microbial growth, and can consequently influence their population demographics, diversity and biological activity [4].

OPTIMIZATION OF BIOSORPTION

Significant studies have exploited the biosorption ability of different bacterial biosorbents against Cr(VI) from the aqueous waste environment. Various physiochemical parameters affect the ability of chromium ions to be absorbed from the environment. It is very much essential to understand the effect of various factors that affect the biosorption process which could provide a clear view on their impact and further help to design the process to achieve a maximum metal ion removal ability. The understanding of these parameters also aids in the wise utilization of biosorbent which can be further reused and recycled efficiently.

A reversible and rapid process, biosorption relies on the specific functional groups of surface-attached biomass to bind ions in an aqueous solution. Compared to other bioremediation processes, the biosorption process is independent of the cellular metabolism and can be used against any type of metal removal, where the latter requires a specific pH based on the microbial growth, whereas the former is used in a wide pH range (1-9) without compromising the effectiveness [11]. Similarly, temperature can also be employed as it is non-metabolic and desorption can be achieved within a short span of time. The major advantage of the biosorption process is cost-effective, where inexpensive agricultural waste and industrial wastes of bio-origin such as residual biomass can also be employed [12]. Another important feature of the biosorption process includes biosorbent regeneration, recovery of metal ions and there is no need of any additional nutrient to perform this process [13].

According to several studies, pH has an impact on the charge of the functional groups on the biomass surface, which is then reflected in the biosorption process. In light of the pH-dependent nature of the biosorption process, researchers speculate that metal ions may bind to the biosorbent through an ion exchange process [14]. In contrast, other studies have concluded that metal ions bind to the biosorbent primarily through functional groups within the cell wall of biomass such as carboxyls, amides, and hydroxyls. In another study, the maximum biosorption was observed at pH 4, which is in accordance with Lin *et al.* [15] argument that hydrogen ions at concentrations greater than 4 deprotonate carboxyl groups in the cell wall, facilitating metal uptake.

When the pH was maintained at lower values, transfer of H⁺ ions from the functional group has indicated that the binding sites for the metal ions were occupied. Additionally, when the pH increases, the H⁺ ion concentration decreases, which leads to negatively charged metal ions binding to positively charged biomass surfaces [11].

Using *S. cerevisiae* as a model for studying the effects of temperature on hexavalent chromium metal ions biosorption, Goyal *et al.* [16] found that increasing the temperature from 25 to 45 °C enhances *S. cerevisiae* capacity to absorb metal ions through higher affinity for metal ions or to increase binding sites for metal ions on the biomass surface. However, at very high temperatures, the metal adsorption tends to be decreased as a result of the distortion of binding sites on the surface of the cell [17,18]. Sepehr *et al.* [19] have also found that temperature has a major effect on the fungal biosorption which influences the enzymatic systems as well as the metal solubility in the effluents and its adsorption rate. Furthermore, Yu *et al.* [20] found that growing temperatures above a certain limit may also affect the integrity of cell membranes, ionization of chemical compounds in biomass cell walls, as well as the stability of metal complexes.

Dönmez *et al.* [21] and Chojnacka *et al.* [22] found that when the concentration of biomass increases, the absorption of metal ions on the surface of biosorbent found to be high thus resulting in increased adsorption yield; the number of active sites may, however, decrease in some cases if partial cell aggregation is observed at higher concentrations [23].

A study by Al-Qodah [24] concluded that an increase in agitation speed decreases the biosorption of heavy metals. Consequently, the thickness of external mass transfer resistance over the applied layer decreased when the agitation rate was increased. As well, as the rate of mixing increases with increasing agitation speed, there is insufficient contact between the binding sites of biosorbent and the metal ions, resulting in a decrease in biosorption efficiency [25].

CHARACTERIZATION OF BIOSORBENTS

Certain sophisticated analytical tools are used to investigate the active sites responsible for metal pollution binding. For instance, Fourier transformed infrared spectroscopy (FTIR) helps to locate the binding sites of the biosorbents and the Scanning Electron Microscopic (SEM) technique helps to visually observe and confirm the surface morphology of the biosorbents [26].

Scanning electron microscopic analysis from various studies shows that untreated bacterial biomass displays smooth cell surfaces. In contrast, hexavalent chromium-treated biomass exhibits high roughness and surface depressions that can be attributed to chromium adsorption on the cell surface. The SEM

micrographs result in a better understanding of cell surface morphology prior to and after biosorption [27].

The FTIR analysis is performed after mixing bacterial biomass with 10 mL of Cr(VI) solution (100 mg/L) at pH 2.0. In the final step of absorption, the pellets are separated, washed twice with double-deionized water followed by drying in an air oven dryer. Additionally, each of the dried Cr(VI)-treated and untreated biomass samples is ground using a mortar and pestle using salts (KBr).

After the powder is obtained, a 7 mm separation disc is produced by manually pressing it. Finally, the IR absorbance spectrum is recorded within the range of 500 to 4000 cm^{-1} . The FTIR analysis of chromium treated and unexposed biomass revealed that different absorption bands appeared at different wavelengths, indicating numerous functional groups. These included amines, hydroxyl groups (bonded and non-bonded), amides, carboxyls, aromatic rings, and alkenes and aliphatic hydrocarbons groups, etc. There were slight differences in peak intensity between the chromium treated and untreated biomass. There were also a few minor peaks that switched.

For unexposed biomass, strong absorption bands were detected at 3299.65 cm^{-1} , indicating that amine N-H stretches are present or that hydroxyl groups (O-H) may be present in that region. Both types of biomass showed similar absorption peaks of aliphatic (-CH₂) groups at 2925 cm^{-1} and 2925 cm^{-1} indicate that lipids, proteins, nucleic acids, and polysaccharides are stretched asymmetrically within the cell wall of biomass samples. These observations led to the conclusion that the changes observed were caused by chromium interacting with the organic functional groups [28].

In addition, Zhang *et al.* [29] have determined how the functional moiety of the dead biomass of the *Corynebacterium* strain SH09 changes during the process of biosorption using FTIR spectroscopy. Metal ions shifted several functional groups in the treated biomass, including amide, particularly C=O, C-N, and ionized carboxyl, respectively, corresponding to spectral peaks observed in FTIR spectrum, 1537 cm^{-1} , 1651 cm^{-1} , and 1385 cm^{-1} , thus suggesting their involvement in biosorption.

As a result of these studies, it appears that the carboxyl, carbonyl and secondary amine functional groups in fungi have played a crucial role in the biosorption process. Many researchers have reported that metal-ion treated biosorbent undergoes group shifts or changes during biosorption of heavy metals [30, 31, 32]. Biosorbents are characterized through scanning electron microscopy, one of the most important and widely used characterization techniques. According to Salunkhe *et al.* [31], metal ions have been detected on the surface of the *C. lunatus* cell biomass during the biosorption process. Several other researchers, Salvadori *et al.* [33], Mishra *et al.* [34], Kim *et al.* [35] have also used scanning electron microscopy to investigate and characterize the presence of metal ions at the surface of cell biomass.

MECHANISM OF BIOSORPTION

Researchers have investigated how bacterial biosorbents play a role in chromium biosorption. It is very essential to understand the mechanisms of metal removal from the environment which enables to develop large scale technology. The biosorption mechanism of hexavalent chromium was demonstrated to be a complex process that was influenced by various factors including, metal properties and structure, nature and structural components of the biosorbent, and also environmental conditions which play a crucial role in the biosorption process.

The common biosorption mechanism found in the removal of chromium metals includes metal chelating, ion exchange, metal adsorption on the surface, microprecipitation, metal diffusion through the cell walls of bacterial biosorbent and on to the membrane using membrane bound proteins. Furthermore, studies reveal that the major forces behind metal's interaction with bacteria include electrostatic interactions, van der Waals forces, and covalent bonds, all of which play a part in binding metal to bacterial biosorbents. During the absorption of metal ions, the bacterial biosorbent has various functional groups that tend to interact electrostatically, including amines, carboxylic acids, and phosphoryl groups in addition to hydroxyl groups [9, 27].

Copper and lead ions are biosorbed by *Bacillus* sp. using an ion exchange mechanism (ATS-1) has been described by Çabuk and colleagues [36]. In the study Kalola and Desai [37], carboxyl and amide groups were found to be crucial for biosorption of *Bacillus subtilis* SS-1 isolated from electroplating industry.

Sharma and Singh [38] remarked that Gram positive bacteria are primarily composed of peptidoglycan and teichoic acid, which are basically acetylglucosamine, acetylmuramic acid, and copryranosyl glycerolphosphate, respectively. These functional groups, such as carboxyl, phosphate, and hydroxyl, play an important role in the formation of bonds between biomass and metals [39]. The presence of lipopolysaccharides, proteins, and phospholipids in the outer surface of Gram negative bacteria blocks the displacement of phosphates and carboxyl groups and provides a favorable site for metal ion binding, according to Cheung and Gu [40].

RECENT DEVELOPMENTS

The two main phenomena towards the development of biosorption procedures are making use of the hybrid technology for the elimination of pollution, in particular using living cells and the advancement of biomaterial immobilization and optimization together with reuse and recycling. Furthermore, the market environment plays a critical role in the development of biosorption technology [27].

Biosorption procedures are successfully used for the treatment of industrial waste waters but they can also be used to serve other purposes. They can blend in various environmental applications where elimination of metal ions is necessary. Substantial research may also be conducted in conventional green roofs. Biosorption technology can be implemented to treat the green roof run off which contains moderate levels of metal pollutants. Inclusion of biomaterials such as seaweeds and crab shells in the green roof setup will serve the function of conventional fertilizers as well as biosorbents for heavy metal ions. Nonetheless, further research findings on biosorbents such as their mode of application, assessment of life cycle and controlled leaching of light metal ions has to be studied [41].

Similarly, the process can be explored to treat heavy metals obtained from comparatively dilute solutions such as effluents released from lab wastes. Given the perplexity of the wastes generated from laboratories, not much emphasis is given to their treatment. Laboratory wastes are considered as potential sources of suitable investigation fields to investigate the application of biosorption treatment machinery, and there is no need to remove metal ions when eliminating contaminants is imperative. Additionally, biosorption can be used to eliminate other types of pollutants as well. They must not be confined to certain areas and pollutants to perform control actions. With a clear primary knowledge of the science governing biosorption and a persistent technological survey, biosorption technology can be applied to non-confined areas and sources of water pollution. Biosorption can be applied to carry out the best managing practices (BMP) effectively. One such example is the development of storm water BMP's which have been implemented in the previous years [19].

Biosorbents can be engaged in the purification of ion-based pharmaceuticals such as proteins, antibodies, and peptides. In order to obtain fine and pure products column chromatographic techniques need to be utilized instead of fixed or moving bed adsorption techniques. Other factors that need to be considered in pharmaceutical functions include consideration of the heat reactivity of biosorbents and monitoring the impurities released during autoclave sterilization and purification. The difficulties faced during biosorption usage act as driving forces by motivating researchers to apply other technologies like hybrid technology which integrates a range of methods for processing of waste water at large-scale [26].

Taking a great step forward in the development of sorbents is essential if we are to solve the significant problem of removing and recovering metal ions. The development of state-of-the-art biosorbents with maximum exclusion ability, however, is necessary to eradicate the toxic effects of metal ions in aqueous solutions. Since, biosorbents are biodegradable in nature, it might be challenging to store them and recycling is mandatory for cost effectiveness and remediation of the environment. Crystalline domains of cellulose are reported to lend strength, resistance to swelling and fibre stability. Therefore, understanding the chemical structure of biosorbents can help us develop cost-effective modifications and improve the removal of toxic metals from aqueous solutions [7].

CONCLUSION

Biosorbents made of bacterial biomass exhibit competencies promising the removal of metal ions from solutions. The bacterial biomasses display a budding potential to contend with standard traditional technologies to treat toxic metal contaminated solutions. Such biomasses are cost-effective and are prepared from inexpensive raw materials that are eco-friendly as well. The one major drawback of this technology is the confinement of the range of research performed to laboratory conditions without really tackling the actual issues. With the existing knowledge on the key theory impacting the performance, mechanism, operation mode, capacity and relative experimental parameters, new progress has to be made in order to convert and apply this technique as a tool to scale-up and decode the potential employability of biosorbents leading to their adequate commercialization.

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CONFLICT OF INTEREST

The authors declare no conflict of interests.

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