Heavy metal evacuation by cyanobacteria

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Abstract

Many pollutants are continuously released from various sources. Among these pollutants, heavy metals adversely affect ecosystems due to their non-degradable nature. These heavy metals remain present in food chain of organisms and negatively affect their health. There are various organisms who show the high potential of eradication of these metals from the environment. Out of these organisms, cyanobacteria which is a group of prokaryotic photo-trophic organisms shown the highest bio-remediation activity from their surrounding due to their remarkable adaptability and versatility. Cyanobacteria have natural capability to produce extracellular polysaccharide and also have several metal binding sites, which have made them a promising candidate for heavy metal removal through biosorption strategy.

Keywords: Biosorption, Extracellular polysaccharides, Cyanobacteria, Heavy metals

Introduction

"The heavy metals can be defined as elements with an atomic number larger than 20 and an atomic density greater than 5gm cm3, along with having the characteristics of a metal. According to United States Environmental Protection Agency, most common heavy metals in order of abundance are lead, copper and Mercury" (USEPA, 1996).

Consequences of heavy metal uptake/contamination

"Heavy metal pollution is one of the major environmental problems that causes serious health issues" (Aktan *et al.*, 2013) [4]. Though certain these metals, *viz*. Copper, Maganese, Iron, Zinc are essential for human health and wellbeing but can become toxic when they reached to higher concentration. "The higher concentration of heavy metals deteriorates the biological system by damaging cellular component specially involves DNA and protein damage, which is going to lead cancer" (Tchounwou *et al.*, 2012) [57].

Methods of evacuation

There are various strategies to evacuate heavy metals from the environment. There are two methods of evacuation, first is physical and another is chemical method. Physical method in which techniques such as ion exchange, reverse osmosis and filtration are involved. While in chemical method, techniques like precipitation, electrochemical treatment and oxidation and reduction are involved.

But all these methodologies exhibit few drawbacks like inadequate evacuation of metals, increase in cost and energy, generation of sludge and useless matter. Hence a perfect cleansing process is required. "To overcome these disadvantages, there is a need for an effective and economical method of evacuation of heavy metals from different contaminants, have resulted in the development of alternative separation technologies" (Zhao *et al.*, 2015) [64].

Hence, there is a need of a strategy which involve living organisms. This type of strategy going to be the best substitute for all existing methodologies of heavy metal eradication. In this regard, biosorption provide good potential for the process of metal eradication. "Biosorption can be defined as the ability of biological material to accumulate heavy metals from wastewater through metabolically mediated or physicochemical pathways of uptake" (Fourest & Roux, 1992). "Some advantages of biosorption can be summarized as follows" (Michalak *et al.*, 2013) [34]

- Absence of toxicity limitations
- Easy absorbance and recovery of biosorbed metals
- Easy regeneration and reuse of biomass
- The possibility of easy immobilization of dead cells
- Avoidance of the sudden death of the biomass population
- Easy mathematical modelling of metal uptake reactors" (Michalak *et al.*2013) [34].

Microorganism involved in heavy metal evacuation

"There are many types of microorganisms involved in the biosorption process, which include bacteria, fungi, yeast and algae" (Kaushik & Malik, 2009) [28]. Out of such microorganisms, cyanobacteria play a very important role. "Cyanobacteria (Blue-green algae) are a morphologically diverse and widely distributed group of photoautotrophic or photosynthetic Gram-negative prokaryotes which exhibit oxygenic (O2-evolving) photosynthesis similar to plants"(Fogg *et al.*, 1973; Stanier & Cohen-Bazire, 1977) [54]. "Cyanobacteria comprise about 150 genera with more than 2000 species" (Van den Hoek *et al.*, 1995) [60]. "Owing to their remarkable adaptability to varying environmental conditions, they successfully colonize almost all kinds of terrestrial (agricultural soils, grassland soils, desert soils, forest soils and rocks), freshwater (rivers, lakes, streams, ponds) and marine habitats" (Tandeau de Marsac & Houmard, 1993). "They possess effective protective and tolerance mechanisms against various abiotic stresses, such as desiccation" (Dadheech, 2010 [12]; Potts, 1999) [42], salinity (Hagemann, 2011 [25]; Klähn & Hagemann, 2010), ultraviolet radiation (Ehling-Schulz & Scherer, 1999 [18]; Quesada & Vincent, 1997) [45], high light intensity (Donkor & Häder, 1995 [17]; Lakatos *et al.,* 2001) [32], extremes of temperature (Hossain & Nakamoto, 2002 [26]; Singh *et al.,* 2005 [53]; Tang & Vincent, 1999), oxidative (Hossain & Nakamoto, 2003) [27], acid (Gopalaswamy *et al.,* 2007) and "heavy metals" (Turner & Robinson, 1995). "Cyanobacteria are equipped with well-developed sensors and signal transducers that perceive and transduce signals from a changing environment or abiotic stresses" (Los *et al.,* 2010) [33]. "Photosynthesis in cyanobacteria saturates at low light intensity. They have a high affinity for light, and maintenance requirements are extremely low" (Van Liere & Mur, 1979) [61]. "Moreover, cyanobacteria are characterized by the low state of cellular differentiation, simple thallus organization, copious production of mucilage (by many genera and species), absence of motile cells, and the presence of chlorophyll *a*, β-carotene, myxoxanthin, myxoxanthophyll and phycobilins or phycobiliproteins (C-phycocyanin, allophycocyanin and C-phycoerythrin) as photosynthetic pigments" (Carr & Whitton, 1982 [8]; Prescott, 1969) [44]. "They are known to synthesize and accumulate a variety of storage compounds (inclusion bodies), such as cyanophycin (a protein-like polymer consisting of equimolar amounts of arginine and aspartic acid), myxophycean starch, glycogen, poly-β-hydroxybutyrate and polyphosphate" (Allen, 1984 [5]; Kromkamp, 1987) [31]. "The biotechnological applications of cyanobacteria in diverse areas, such as bioremediation and pollution control, bioenergy and biofuels, and nutraceuticals, have been well-documented" (Patterson, 1996 [39]; Abed *et al*., 2009 [1]; De Phillips *et al*., 2003 [14]; Pandey, 2010) [38].

"Several cyanobacteria exhibit effective mechanisms for heavy metal tolerance or resistance up to certain concentrations, employing various strategies such as metal efflux as well as production of metallothioneins (cysteine- rich metal-binding proteins) and extracellular polysaccharides" (Sandau *et al.*, 1996; Turner and Robinson, 1995 [58, 59]; De Phillips *et al*., 2003 [14]; Robinson *et al*., 2000; Pereira *et al*., 2011) [40]. "Cyanobacterial extracellular polysaccharides (EPS) are heteropolymers and possess distinct characteristics unlike other bacteria. It includes highly anionic nature resulting from uronic acids, sulphated sugars and a diverse composition of monosaccharides. But the role of EPS in metal remediation is due to the presence of negatively charged groups such as sulphate, phosphate, carboxyl and hydroxyl groups. These groups may work as chelating agents for positively charged metals" (De Philippis & Micheletti, 2017; Cui *et al*., 2021 [10]; Potnis *et al.*, 2021 [41]; Bhatt *et al.*, 2022; Ciani & Adessi, 2023) [9].

There have been several studies where cyanobacteria were used for evacuation of heavy metal by the technique of biosorption. Sharma *et al.*, 2008 [50] "worked on the sequestration studies of chromium from dilute aqueous solutions using the exopolysaccharide-producing cyanobacteria *Nostoc* and *Gloeocapsa" (Sharma et al,2008)* [50]. Shukla *et al*., (2012) [52] "studied the chromium (CrVI) removing potential of a cyanobacterial mat (consortium of *Chlorella* sp. *Phormidium* sp., and *Oscillatoria* sp.) which was collected from the effluent site itself" (Shukla *et al*., 2012) [52]. "Cyanobacteria-mediated biosorption continues even after the cyanobacteria die; in other words, the dead cells of cyanobacteria can also be employed for the biosorption of heavy metals" (Gupta *et al*., 2015) [24]. Cyanobacteria exhibit a high metal binding activity and such a characteristic makes this organism an efficient biosorbent microorganism.

"Several cyanobacteria genera, such as *Anabaena, Cyanobium, Nostoc, Cyanothece, Arthrospira, Microcystis, Synechocystis,* and *Leptolyngbya* have shown promising results on Cu, Cd, Zn, Cr, Pb, Ni, Co or Hg removal with initial concentration ranging from some mg/l to 150-200mg/L" (Mota *et al*., 2016; Zinicovscaia *et al*., 2018 [65]; Yadav *et al*., 2021 [63]; Bloch & Ghosh, 2022, Pandey *et al*., 2022) [37]. "Maximum uptake is typically in the range of 15-80mg/g dry weight, but some works have presented values even higher than 300mg/g dry weight" (Cui *et al*., 2021) [10]. "Also, the use of consortia of different cyanobacteria species or microalgae / other microbes and cyanobacteria may help to attain higher metal tolerance as well as higher metabolite synthesis, positively contributing to metal removal" (Cui *et al*., 2021). "Generally, since the removal efficiency is maximized with a lower initial metal concentration, biosorption or bioaccumulation by cyanobacteria can be adopted after conventional methods that are characterized by low efficiency at a low metal concentration"(Agarwal *et al.*, 2020) [2]. "Diengdoh *et al.*, 2017 [16] isolated *Nostoc muscorum* and studied its potential for Zn+2 removal from aqueous medium" (Diengdoh *et al.*, 2017) [16]. "Mota *et al*., 2016 [35] investigated the removal of Pb+2, Cd+2 and Cu+2 by *Cyanothece* sp. CCY0110. Results exhibited that *Cyanothece* sp. has high removal efficiency for heavy metals" (Mota *et al*., 2016) [35]. "The study was aimed at assessing the biosorption of heavy metals (cadmium, mercury and lead) using the living/dead cells of *Oscillatoria limosa"* (Sivakami *et al*., 2015). "Results indicate that the order of uptake of Cd, Hg and Pb was found to be the order of 82, 78 and 72%, respectively. The study also indicated that the metal uptake appeared to be a concentration independent phenomenon where an increase in metal concentration resulted in an increased uptake of metal" (Cyriac, 2020) [11].

Biosorption through cyanobacteria

"The whole process of heavy metal ion binding to the cell wall may be metabolism dependent or independent, as shown in Figure 1" (Volesky *et al*., 1993) [62]. "In a metabolism-independent process, physiological interaction between the metal and the functional groups like polysaccharides, proteins and lipids has several metal binding groups such as carboxyl, sulphate, phosphate and amino group present on the cell surface. These are used for metal uptake, and this type of mechanism is rapid and can be reversible" (Sardrood *et al.*, 2013).

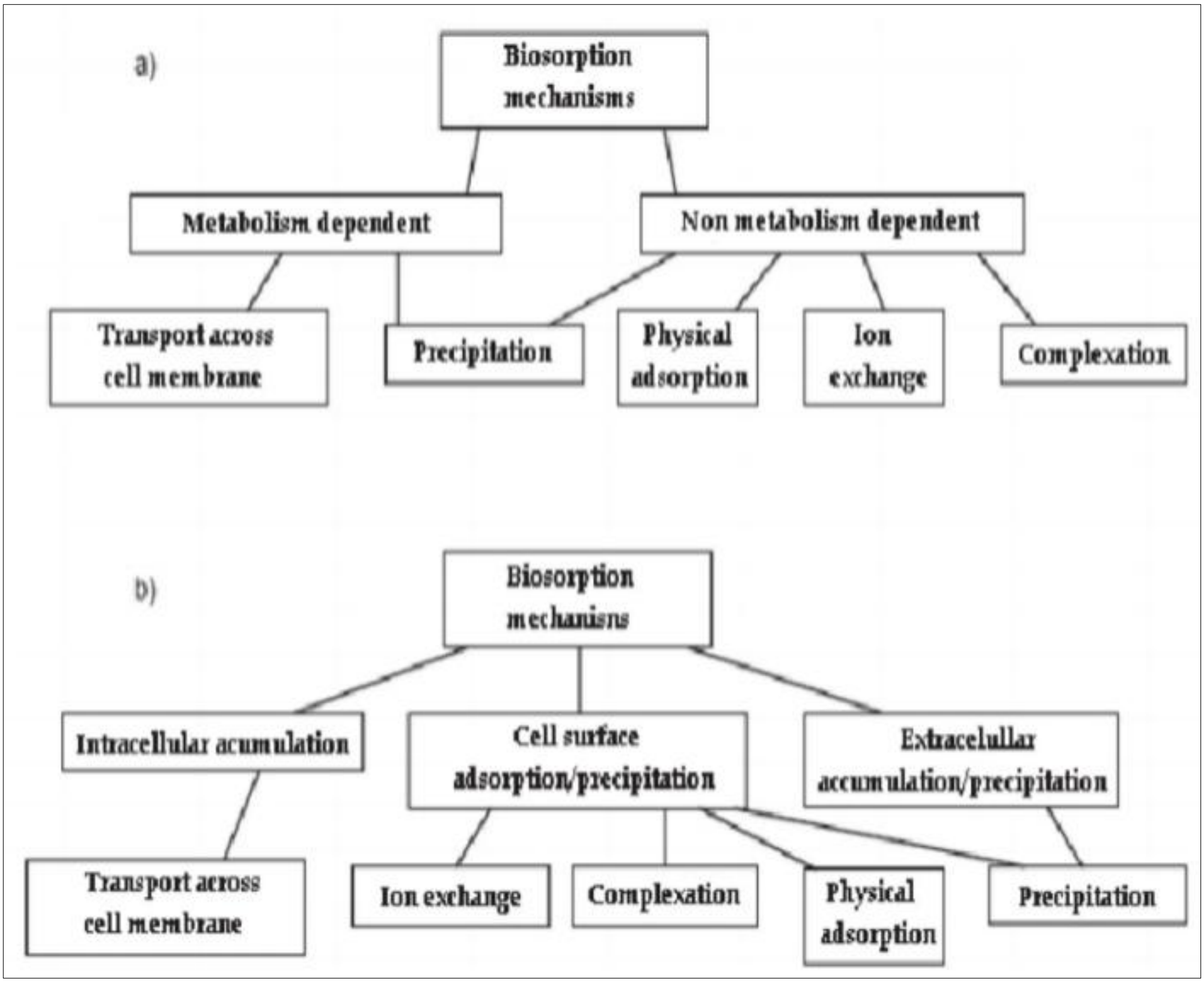

Fig 1: Biosorption mechanisms

The transfer of heavy metal across the cell membrane of microorganisms take place in the same manner as the transport of ions like K+, Mg+ and Na+ occur inside the cell. Hence, in spite of essential ions; the microbial nutrient system by mistake absorbs the heavy metal ions due to their structural similarity.

There is a need of a high-quality biological material that binds the pollutants for the effective strategy of biosorption. The very first step in this process is choosing the right biological material (like algae, fungi or plant) that can clean up the pollutants effectively. Afterwards the treatment of biomass with chemical or heat occur to expose its binding sites followed by the fixing the loose biomass into solid beads or a matrix which prevent it from washing away. It determines faster trapping of heavy metals. This whole experiment done by batch and column method and lastly binding metal ions are extracted by means of desorption which is a reverse process of biosorption where a chemical solution is used to wash off and recover the trapped metal. The biological material i.e. biomass leftover after the procedure can be used again which is a cost effective approach.

Factors affecting the process of biosorption in cyanobacteria

Temperature

"The optimum temperature for biosorption efficiency is within the range of 20-35 °C" (Aksu & Donmez, 2001) [3]. "The exothermic nature of some biosorption processes cause an increase in temperature around 50 °C; this may cause permanent damage to living cells, which in turn decreases the metal uptake process" (Oyewole *et al.*, 2018).

Effect of pH

"The pH of the solution may play an important role in the biosorption process because it affects the nature of biomass binding sites and metal solubility. It also influences the solution chemistry of the metals, the activity of functional groups in the biomass and competition of metallic ions" (Deng & Wang, 2012). The change in pH brings an alteration in the activity of metal binding sites. The eradication of metal ions remains minimum and negligible in the range of pH 2; but as pH reaches from 3.0-5.0 an increase in metal uptake is found. Thus, there is a need of optimization of pH to achieve the maximum eradication of metals.

Effect of biomass concentration

"Biomass concentration is of great importance for the specific metal uptake; for lower values of biomass concentration, there is an increase in the specific uptake. High biomass concentration restricts the access of metal ions to the binding sites" (Gadd, 2010) [21]. "Besides, many physical and chemical pre-treatment can affect permeability and make the metal binding group accessible for binding, which may increase the amount of metal uptake" (Oyewole *et al.*, 2018) [36].

Advantages of the biosorption process

"The process of biosorption was naturally rich, renewable biomaterial, which can be cheaply produced; it also deals with low capital investment and low operational cost, and it has the ability to handle multiple heavy metals and mixed wastes" (Aktan *et al.*, 2013) [4]. "Furthermore, this process has improved the recovery of bound heavy metals from the biomass and also reduced the level of hazardous waste produced" (Aksu & Donmez, 2001) [3].

Future perspectives

The technique of biosorption through cyanobacteria is a sustainable and eco-friendly approach which bring about a double out come in terms of cost and energy."The recovery of nutrients such as C, N, P and others, as well as cyanobacteria biomass production also achieved" (Sachdeva *et al.*, 2018 [47]; Gomes Gradissimo *et al.*, 2020; Kholssi *et al.*, 2021 [29]; Prabha *et al.*, 2022). "Hence, the cost associated with artificial salts and water requirements is reduced, together with the environmental footprint of the process" (Sachdeva *et al.*, 2018).

Conclusion

Cyanobacteria are highly versatile, adaptable, and diverse group of gram-negative phototrophic prokaryotes. The outer surface of cyanobacteria comprises polysaccharide which is called as extracellular polysaccharide. These polysaccharides contain various sites for binding of organic and inorganic pollutants. Cyanobacteria may act as an excellent biological tool for removing heavy metals from their surroundings. However, there is a requirement of extensive research for standardization of the technique of biosorption at an industrial level.

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