

## STUDY OF BIOSORPTION OF HEAVY METALS USING AGRICULTURAL AND MICROBIAL BIOMASS: A REVIEW

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### ABSTRACT

With increasing urbanization and industrialization, pollution is becoming a severe problem. Industries discharge a large amount of untreated or partially treated water into water bodies which contain a variety of harmful effluents in them. Waste water discharged from industries also contain heavy metals like mercury, lead, cadmium, chromium(VI). These metal ions being non biodegradable are lethal for all forms of life, enter the food chain and start accumulating especially in higher trophic levels. Various techniques are available for recovery of metals but due to high operational and maintenance costs, production of large amounts of toxic sludge these methods are inefficient and economically unviable. Biosorption of heavy metals by agricultural and microbial biomass can emerge as a promising alternative for removal of heavy metals from aqueous solution by binding with metal ions. This paper makes an effort to review the variety of agricultural and microbial biomass available for biosorption of heavy metals thus controlling water pollution.

**KEY WORDS** : Biosorption, heavy metals, agricultural biomass, microbial biomass

### INTRODUCTION

Population explosion, urbanisation and industrialization have resulted in discharge of a large amount of pollutants in the water thus leading to deterioration of water quality (Bhatnagar *et al.*, 2010). Effluents which pollute water can be of two kinds-Non point source of pollution which enters the water from scattered locations. Point sources are the effluents being discharged from industries where the source of pollution is known and may consist of volatile organic compounds, waste from food processing industries, various dyes and heavy metals (Abdi *et al.*, 2015). Chosen for their durability, strength and resistance to changing weather metals are used in a number of industries like construction, dentistry, electroplating and mining. These heavy metal ions when enter the environment, especially water are a issue of serious concern as they can travel from one trophic level to another trophic level through food chains and can accumulate in tissues of higher organisms which

can result in severe health problems (Volesky, 2001).

### MATERIALS AND METHODS

Agricultural wastes containing rice husk, rice straw, wheat bran, Bengal gram husk, tur dal, tamarind husk, sugarcane bagasse, cauliflower wastes and barley straw.

Microbial mass containing algae, bacteria and fungi were the biosorbents for the removal of heavy metal ions from water.

#### Conventional methods for the recovery of heavy metals are:

- Chemical precipitation (Murnane *et al.*, 2018) is the most common method for removal of toxic heavy metals from water by addition of counter ions to reduce the solubility of metal ions so that metal ions can precipitate by forming sulphides, hydroxides, phosphates and carbonates. It is primarily used for the removal of metallic cations but can also be used for removal of anions.

- Reverse Osmosis (Shafiq *et al.*, 2018) is a method which can remove metal ions from water by pushing the water under pressure through a semi permeable membrane.
- Membrane filtration process (Serrano *et al.*, 2010) uses semi permeable membrane of different pore size which allows the water to pass through the membrane and retain the metal ions.
- Electrocoagulation (Qdais, 2004) is an electrolytic cell which uses electric current to remove metals from solution. In this method ions are neutralized by ions of opposite electric charge, where they destabilize and precipitate in a stable form to form a floc and can be removed easily.
- Ion exchange method (Khulbe *et al.*, 2018) can remove heavy metals by using natural or synthetic resins by exchanging cations or anions from surrounding metal ions. These resins can be regenerated and used again for removal of metal ions.

All the techniques which have been mentioned above are expensive due to high operational, maintenance costs and also toxic sludge generation (Shamim, 2018).

## RESULTS AND DISCUSSION

### Biosorption

Biosorption is an alternative method for the removal of heavy metal ions from aqueous solutions using biological materials. It is a physicochemical process that occurs naturally and allows binding of desired contaminants generally metal ions by biological biosorbents (Michalak *et al.*, 2013). Biological biosorbents include agricultural biomass and various microbes. Biosorption is defined as the property of certain types of dead, inactive and microbial biomass to accumulate heavy metals from aqueous solutions (Fard *et al.*, 2011) It is a process through which metal ions bind to various functional groups present on the Biosorbent. Biosorption process consists of a solid phase which is the Biosorbent and the liquid phase which is the solvent generally water containing dissolved substances (sorbate/ metal ions). The main advantages of this process include it is cheap to operate (Ahalya, 2003), simple to operate (Wang, 2009), less quantity of sludge generation (Abbas, 2014), high efficiency, can show regeneration of adsorbent and no additional nutrients are required (Nguyen, 2013).

### Factors affecting biosorption

- Temperature (Rosales, 2012) The optimum temperature required for biosorption to occur is between 20-35. Higher temperature above 45 can reduce the biosorption efficiency of biomass.
- pH- pH is an important parameter which affects the rate of biosorption. The optimum PH range for biosorption to occur efficiently is between 3-6, higher pH can decrease the rate of biomass Biosorption (Priya, 2012).
- Biomass concentration- Biomass concentration is directly proportional to uptake of metal. At low cell density more metal ions are absorbed as compared to high cell density.
- Affinity of heavy metal to biosorbent- Certain physical and chemical treatments like treatment of biomass with acid, alkali and heat can alter and bring structural changes in biomass which can make the binding of metal ions easier to biosorbents (Saha, 2019).

### Mechanism of Biosorption

Mechanisms of biosorption are complicated reactions which are not still completely understood. Depending upon the cell's metabolism- biosorption can be classified as metabolism dependent mechanism which is generally seen in living biomass or metabolic independent pathway seen in inactive biomass. Biosorbents contain a large number of functional groups like carboxyl, amine, hydroxyl, phosphate and sulfhydryl groups that can attract and sequester metal ions. Biosorption can include many passive (Salman, 2015), non metabolic mechanisms :

- Complexation (Mahamadi, 2011) - which involves formation of a complex of metal ions to compete with protons for organic binding sites. Complexation can be electrostatic or covalent. Hard metals like K, Ca, Mg, Na bind to oxygen containing hard ligands such as hydroxyl, carbonate and carboxyl. Soft metals (precious metals Au, Ag Pt bind covalently to the cell wall by soft ligands containing nitrogen and sulphur.
- Chelation- which is a type of bonding of ions and molecules to metal ions to form a ring structure which involves the presence and formation of two or more coordinate bonds between a single central atom and a polydentate ligand which is also called as chelant.
- Coordination- The reaction of one or more ligands with a metal ion to form a coordination

compound. It is a reversible reaction consisting of oxidation and reduction. The atom within a ligand that bonds to central atom is called the donor atom. Not very precise information on the theory of coordination chemistry is available for metal absorption but metal species chemical coordination and the stereochemical characteristics are important for their attachment on the available ligands by microbial cell.

- Ion exchange- are the reactions which involve exchange of metal ions with the counter ions present on biosorbent surface. Ion exchange can take place by anion or cation exchange. Microorganisms cell wall is made up of polysaccharides and divalent metal ions exchange with polysaccharide counter ions (Rungrodnimitchai, 2014).
- Precipitation- It can be dependent or independent on the cellular metabolism. When precipitation is dependent on cellular metabolism, metal ions removed are associated with the defense system of organisms and in the latter case precipitation can be the result of chemical reaction between the metal and cell surface (Javanbhakt, 2014).
- Reduction- Metal ions like Au and Pd first get bound to carboxyl group within the cell wall and then these sites become nucleation points for metal reduction and metals can be reduced once they get attached to biosorbent.

### Type of Biosorbents

In the past few decades, the scientific and engineering community has shown interest in sequestering metal ions by living or inactive biomass (Volesky, 1995). Biosorption by biomass waste is seen as a promising environment friendly

technique for recovery of heavy metals from aqueous solutions (Inoue *et al.*, 2017). A wide variety of biomaterial found in nature can be used as biosorbent for the removal of heavy metal ions. Agricultural and microbial biomass can be used efficiently to remove heavy metal ions (Qdais *et al.*, 2004). Recent biosorption experiments have diverted attention on use of agricultural waste material like rice husk (Negm *et al.*, 2017), wheat bran (Das *et al.*, 2019), sugarcane Bagasse (Sarker *et al.*, 2017) for heavy metal removal. Apart from agricultural biosorbents, microbes especially algae (Ali *et al.*, 2018), bacteria (Vijayaraghavan *et al.*, 2015) and fungi (Cerimi *et al.*, 2019) have also drawn attention for the biosorption of metal ions from aqueous solution.

### Agricultural biomass as biosorbents

Recent researches have shown that agricultural wastes can be used as an efficient biosorbent for removal of heavy metal ions. In India, agricultural waste is readily available which contains cellulose, hemicellulose, lignin and proteins. Agricultural biomass contain many functional groups like amino, phenolic, alcoholic, carbonyl which have an affinity towards metal ions and can combine with them to form metal complexes leading to heavy metal removal, as seen in Table 1.

### Microbial Biosorbents

Microorganisms which include algae, bacteria, yeast and fungi are being used as biosorbents for the removal of metal ions from wastewater. Dead microbial mass over living is preferred for the removal of metal ions due to their non requirement for nutrients and also there is no need to monitor BOD and COD (Ayansina *et al.*, 2017) Microbial

**Table 1.** Certain Heavy metals adsorbed by agricultural residue

Agricultural Biomass	Metal Ion removed	Biosorption Capacity(mg/g)	Reference
Rice Husk	Pb(II)	68.0	(Wong <i>et al.</i> , 2003)
	Ni(II)	51.89	(Shafey, 2010)
	Hg(II)	384	(Dadhich <i>et al.</i> , 2004)
Rice Straw	Cr(VI)	3.14	(Gao <i>et al.</i> , 2008)
Wheat bran	Cu(II)	8.34	(Farooq <i>et al.</i> , 2010)
Bengal gram Husk	Cr(VI)	91.64	(Ahalya <i>et al.</i> , 2005)
Tur Dal Husk	Cr(VI)	66.65	(Ahalya <i>et al.</i> , 2007)
Tamarind Husk	Ni(II)	74.62	(Popuri <i>et al.</i> , 2007)
Sugarcane Bagasse	Ni(II)	2.0	(Aloma <i>et al.</i> , 2011)
Cauliflower Waste	Pb(II)	60.57	(Hossain <i>et al.</i> , 2014)
Barley Straw	Pb(II)	23.20	(Pehlivan <i>et al.</i> , 2009)

biosorbents contain a large number of functional groups which can bind to metal ions thus helping in their removal, as seen in Table 2.

### Algae as biosorbents

Algae are aquatic plants belonging to kingdom protista which show photosynthetic activity. Algae lack true roots, stem, leaves and a vascular system for conduction of water. Table 2. Algae are unicellular and microscopic or can be multicellular and macroscopic. Algae are classified as:

Green algae- (*Chlorella*), Cell wall of green algae is made up of cellulose and polysaccharides which contain functional groups like hydroxyl, sulfhydryl, carboxyl and amino groups.

Brown algae- (*Punctaria*), Cell wall contains alginic acid, cellulose and sulfated polysaccharide which contain carboxyl as the main functional group.

Red algae- (*Mastocarpus stellatus*), Cell wall contains sulfated polysaccharide containing galactans with carboxyl and hydroxyl as the main functional groups

### Bacteria as biosorbents

Bacteria are prokaryotic, microscopic and single celled organisms which lack chlorophyll and are adapted to survive in a variety of environmental

conditions like ocean depths, below large rocks and can even stay in the human digestive tract. Most of the bacteria appear in three shapes- spiral, cylindrical and coccus. Bacteria have a simple cell structure as they lack nucleus as well as other membrane bound organelles. Bacterial DNA is found in a folded mass called a nucleoid. Bacterial cells are surrounded by an outer cell wall and an inner cell membrane, some bacteria for further protection are surrounded by a capsule. Bacteria contain a large number of biosorption sites and have a high surface to volume ratio (Kuppusamy *et al.*, 2008) and can be used efficiently to remove metal ions from wastewater. Metal ions when they come closer near the bacterial cell wall, get attached to the functional group present on the cell wall. Gram positive bacteria are better biosorbents because of the presence of glycoproteins than the gram negative bacteria which contain phospholipids

On the basis of bacterial cell wall composition and the ability to hold the gram stain, bacteria have been divided in two major categories:

Gram positive- Bacteria which do not have an outer membrane and consist of thick peptidoglycan layer and a small amount of polyalcohols and teichoic acid. Gram positive bacteria can retain the crystal violet color and stain purple even when washed with alcohol.

**Table 2.** Metal ions adsorbed by various algal species

Algal Species	Metal Ion removed	Biosorption Capacity(mg/g)	Reference
<i>Spirogyra</i> sp	Pb (II)	45.4	(Gupta <i>et al.</i> , 2008)
	Cu (II)	13.7	(Gupta <i>et al.</i> , 2006)
<i>Sargassum filipendula</i>	Cu (II)	84.10	(Sheng <i>et al.</i> , 2007)
	Ni (II)	62.79	(Mahmood <i>et al.</i> , 2017)
<i>Rhizopus cohnii</i>	Cd (II)	40.5	(Jinming <i>et al.</i> , 2010)
<i>Spirulina platensis</i>	Cu (II)	67.93	(Hadeel <i>et al.</i> , 2014)
<i>Enterobacter</i> sp	Pb (II)	50.32	(Lu <i>et al.</i> , 2006)
	Cu (II)	46.2	
<i>Cladophora</i>	Pb (II)	46.51	(Jafari <i>et al.</i> , 2012)
	Cu (II)	14.71	(Deng <i>et al.</i> , 2007)

**Table 3.** Biosorption of heavy metal ions by bacterial biomass

Bacteria Species	Metal Ion removed	Biosorption Capacity(mg/g)	Reference
<i>Micrococcus</i>	Ni (II)	84.27	(Rani <i>et al.</i> , 2010)
<i>Pseudomonas aeruginosa</i>	Cd (II)	90.41	(Chellaiah, 2018)
<i>Bacillus subtilis</i>	Pb (II)	78.8	(Wierzba, 2015)
<i>Enterobacter</i> sp	Pb (II)	50.9	(Wang <i>et al.</i> , 2009)
<i>Pseudomonas putida</i>	Pb (II)	50.4	(Pardo <i>et al.</i> , 2003)
<i>B.thuringiensis</i>	Ni (II)	45.9	(Ayten, 2007)

**Table 4.** Biosorbents of Heavy metals by Fungi

Yeast Species	Metal Ion removed	Biosorption Capacity(mg/g)	Reference
<i>S. cerevisiae</i>	Hg (II)	76.20	(Wifak <i>et al.</i> , 2017)
	Pb (II)	60.24	
<i>Penicillium chrysogenum</i>	Ni (II)	82.5	(Bahafid <i>et al.</i> , 2017)
<i>P. purpurogenum</i>	Cr (VI)	36.5	(Say <i>et al.</i> , 2004)
<i>Aspergillus niger</i>	Pb (II)	34.4	(Dursun, 2003)
<i>Streptomyces</i> sp	Cr (III)	76	(Saurav <i>et al.</i> , 2011)
	Cr (VI)	84.27	
<i>PhanerochaeteChrysosporium</i>	Ni (II)	55.9	(Ceribasi <i>et al.</i> , 2001)
	Pb (II)	53.6	

Gram negative - Bacteria which have an additional outer membrane made of polysaccharides and phospholipids and consist of a thin layer of peptidoglycan. These are resistant against antibiotics and are often pathogenic. Gram negative bacteria do not retain the stain color when they are washed with alcohol.

#### Fungi as biosorbents

Fungi are eukaryotic organisms which include yeasts, molds and mushrooms that can exist as a filamentous or multicellular body. Fungi contain cells which have nuclei and membrane bound organelle. Earlier fungi were included in plant kingdom but due to lack of chlorophyll in them fungi have been separated from plant kingdom Fungi cell wall is made up of chitin.

Yeasts are eukaryotic, unicellular organisms belonging to the fungi kingdom. Yeast species *Saccharomyces cerevisiae* also known as baker's yeast is greatly used as a biosorbent material for removal of metal ions due to presence of carboxyl, amine and phosphate group present on yeast surface also industrial manufacture of yeast is economical thus making it a cost effective biosorbent. Yeasts are non specific in terms of metal biosorption and thus can bind to a broad range of metal ions, as seen in Table 4.

#### Regeneration of Biosorbents

For the commercial use of agricultural and microbial biosorbents, regeneration of biosorbents is essential for recovery of heavy metals as well as to reduce the cost involved in biosorption. Process of regeneration should make the biosorbent come back to its original condition so that it can be reused along with the recovery of metals (Ismail *et al.*, 2016). Regeneration is done by washing the biosorbent containing heavy metals in dilute

solutions of hydrochloric acid, sulphuric acid, nitric acid and acetic acid (Bakir *et al.*, 2010) of appropriate strength. Biosorbent containing Cd(II) was desorbed using  $\text{CaCl}_2$  and metal containing biosorbent was separated by filtration (Vimala, 2011)

*Saccharomyces cerevisiae* after it had shown biosorption of metal ions was desorbed by dil. Nitric acid and was regenerated by sodium hydroxide and mineral free water (Farhan *et al.*, 2015). *Canna indica* containing metal ions like Cd, Pb and Cr was desorbed with dilute hydrochloric and nitric acid and was reused for removal of heavy metals (Dixit *et al.*, 2015).

#### CONCLUSION

Biosorption is a cheap and eco-friendly method for removing heavy metals from aqueous solution. Despite of being a huge market for green and sustainable technology use of biological biosorbents for metal removal is done only at micro level as these biological biosorbents contain many functional groups present on their surface thus making them non selective for metal ions, this could be overcome by making biological biosorbents specific to particular metal ions. Emphasis on regeneration of biosorbents is also an important issue to retain the economic feasibility of using biosorbents.

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