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Biosorption of Heavy Metals as a New Alternative Method for Wastewater Treatment: A Review

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ABSTRACT

Heavy metals are recognized as the most significant environmental concern, since they are a major source of wastewater pollution. Human activities and industrialization have mostly resulted in the discharge of heavy metal-containing pollutants into water resources, contaminating them and endangering the health of humans and the environment. Many studies on wastewater treatment procedures such as precipitation, evaporation, ion exchange, membrane processes, and electroplating have been done. However, these traditional methods are costly, non-renewable and produce secondary pollutants. We concentrated on biosorption in this review because it is thought to be the most promising alternative strategy for eliminating hazardous metal ions from water sources. Biosorption is a physical process that employs ion exchange, surface complexation and precipitation to use less expensive alternative biological materials as biosorbents. Various biomasses including microorganisms (bacteria and fungi), algae and plant products have been used as biosorbents for metal biosorption. Biosorption with local microbiota has inspired considerable interest in the removal of harmful heavy metals from wastewater without creating any detrimental consequences in recent years. Microorganisms, particularly fungi (both live and dead), have been recognized as a potential class of low-cost adsorbents for heavy metal ion removal in solution. The biosorption behavior of fungal biomass attracts the attention due to its numerous advantages; consequently, additional study is required to completely exploit it in wastewater treatment.

INTRODUCTION

Heavy metal contaminations are a severe worldwide environmental issue since they are non-degradable and not easily accessible for absorption by living organisms **(Taha *et al*., 2023)**. Metal ions such as lead, cadmium, mercury, copper, chromium, zinc, nickel, and cobalt are very toxic to animals, with a huge environmental impact **(Volesky & Holan, 1995)**. If consumed in excess, all heavy metals are poisonous and have negative effects on organisms. Under some conditions, they may be necessary for living organisms' growth and development **(Taha *et al*., 2023)**. The kind of living creature, the dosage and the contact period are all factors that determine heavy metal toxicity **(Taha *et***

***al*., 2023)**. Heavy metal removal procedures (such as precipitation, filtration, ion exchange, carbon adsorption, evaporation, membrane technology, reverse osmosis pre-concentration, redox, electrowinning, chelation, wastewater coagulation, and electrochemical) have all been shown to be ineffective (**Ali *et al*., 2019; Taha *et al*., 2023)**.

For their inadequate removal and unpredictable metal removal, most of these procedures are very costly for large-scale deployment and risky for continual monitoring and control **(Da Rocha Ferreira *et al*., 2019)**. Furthermore, they are impractical and lack specificity for metal-binding characteristics (**Ayangbenro & Babalola, 2017)**. In addition, they eliminate non-target beneficial microbial biota such as nitrogen-fixing bacteria and other fauna species **(Siddiquee *et al*., 2015)**.

The primary emphasis on bioremediation as a green (biological) therapy and economically viable biotechnology **(Wang & Tam, 2019; Taha *et al*., 2023)** is strongly advised to overcome the limitations of traditional approaches **(Joshi, 2018; Taha *et al*., 2023)**. A biological technique also has the benefit of be able to treat a high volume of wastewater while maintaining low biomass content and a short operating period **(Akkar *et al*., 2016)**. Through the activity of microbes and plants, bioremediation includes degrading, eliminating, modifying, immobilizing, or detoxifying different chemicals and physical pollutants from the environment.

Biosorption, biofilters, bioventing, bio-augmentation, biotransformation, composting, land farming, bioreactors and biostimulation are the most well-known bioremediation processes **(Siddiquee *et al*., 2015; Taha *et al*., 2023)**. The biosorption method is a relatively new process that is becoming a potentially preferred alternative method for the decontamination of heavy metal-containing effluents due to its various advantages over traditional methods, such as its low cost, better efficiency, high sensitivity, low technology requirement, minimal chemical/biological sludge production, minimal requirement of additional nutrients. Thus, biosorption is now considered one of the most promising strategies for the sequestration of harmful and toxic metals **(Fu & Wang, 2011)**. Biosorption mechanisms include but are not limited to ion exchange, coordination, complexation, chelation, adsorption, micro-precipitation and diffusion through cell walls and membranes. These mechanisms can change depending on the species used, the origin and processing of the biomass and the solution chemistry **(Churchill *et al*., 1995)**.

Heavy metals are tolerated and accumulated by microorganisms (bacteria, mold, and yeast) and algae. Microorganisms transform, alter and use hazardous contaminants to produce energy and biomass, repairing the ecosystem and avoiding further pollution. Using bacteria as biosorbents to clean up wastewater is an innovative concept **(Goutam *et al.*, 2021; Taha *et al*., 2023)**. Biosorption through various fungal species (live or dead) has received a lot of interest for usage as biosorbents in the elimination of hazardous metal ions **(Saad, 2015)**. The living biomass of metal-binding ions is determined by numerous variables, including the followings: nutrients, environmental circumstances and cell age are all factors to consider **(Kapoor & Viraghavan, 1995)**. However, in high quantities, heavy metals may harm living biomass. As a result, dead biomass is strongly suggested to address the drawbacks of live biomass **(Butter *et al.*, 1998)**.

1. Biosorption

One of these crucial processes, known as biosorption, requires a more strongly bonded solid phase (the biosorbent) and a liquid phase (often water) containing dissolved sorbed material (sorbate: metal ions) **(Dhankhar & Hooda, 2011)**. It is regarded as a quick phenomenon of non-growing passive metal sequestration; this is the process whereby inert biological materials or materials derived from biological sources passively absorb harmful substances.

Several biomaterials, including microorganisms (bacteria, mold, and yeast), algae and plant byproducts have been studied for their biosorption capabilities and indicated significant potential for metal precipitation from water (wastewater or water resources) **(Petersen *et al*., 2005)**. Microorganisms such as bacteria and fungi are thought to be efficient suppliers of biosorbents **(Wang & Chen, 2009)**. These biosorbents have metal-sequestering capabilities and may be utilized to reduce heavy metal ion concentrations in solution from ppm to ppb **(Abbas *et al*., 2014)**. This efficiency is due to the makeup of the cell wall and the functional groups involved in metal binding. Many microorganisms remove heavy metals by surface adsorption, which involves metals accumulating on the cell surface and interacting with functional groups on the cell surface such as carboxyl, amine hydroxyl, phosphate and sulfhydryl groups **(Das *et al*., 2008; Kisielowska *et al*., 2010)**.

Biosorption happens under a range of physiochemical parameters such as temperature, pH and the presence of other ions. Because of its quick kinetics, it can treat enormous amounts of wastewater. Biosorption employs naturally rich renewable biomaterials that are inexpensive to create and reduce the need for extra costly chemicals, which generally cause disposal and space issues **(Zaki *et al*., 2022)**. Biosorption provides excellent selectivity for the recovery and elimination of heavy metals. Furthermore, biosorption has a cheap capital investment and operating cost, and it can handle a variety of mixed wastes and heavy metals **(Aktan *et al*., 2013)**. Finally, biosorption has increased the recovery of bound heavy metals from biomass while decreasing the amount of hazardous waste created **(Aksu & Dönmez, 2001)**. However, biosorption has two major drawbacks: metal desorption needed because of ongoing use regardless of metal value, and early saturation occurring when all metal interaction sites are used up **(Tabaraki *et al*., 2013)**. The biosorbent's adsorption characteristics are uncontrollable by biology and are formed during pre-growth **(Colak *et al*., 2011)**.

2. Biosorption mechanisms

Several methods have been reported for the biosorption-based removal of dangerous chemicals. Metabolism-dependent or -independent categories were established **(Veglio & Beolchini, 1997)**. While, metabolic activities are required for bioaccumulation, biosorption occurs without metabolic involvement **(Volesky, 2007; Chojnacka, 2010)**. In contrast to the active process of bioaccumulation, the passive biosorption process may be completed in a shorter amount of time. Biosorption is a multi-step process that includes adsorption, chelation/complexation, ion exchange and surface precipitation **(Farooq *et al*., 2010; Bilal *et al*., 2018)**. The biosorption mechanisms are shown in Fig. (1) and discussed as follows:

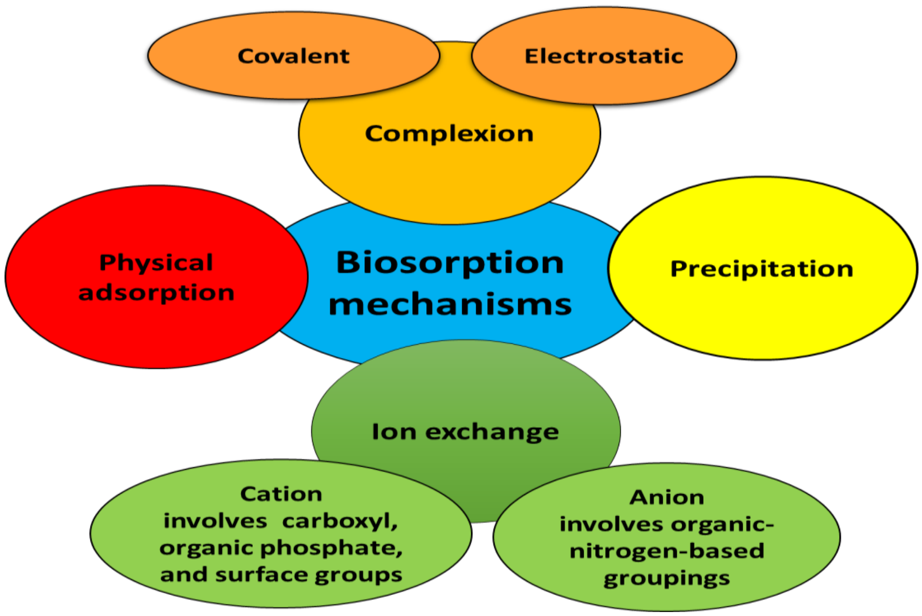

Fig. 1. Schematic diagram involving biomass biosorption mechanisms.

2.1. Ion exchange

Different functional groups on the surface of biomass are hypothesized to facilitate ion exchange as the major mechanism of biosorption. Polysaccharides present in microorganism cell walls are metal ion exchange sites due to the polysaccharide's opposing charge **(Chojnacka, 2010; Vijayaraghavan & Balasubramanian, 2015)**. Cell walls of different species have different compositions; for example, bacterial cell walls are composed of peptidoglycan; fungal cell walls are composed of chitin, and algal cell walls are composed of alginate and sulfonated polysaccharides **(Bilal *et al*., 2018)**.

2.2. Complexation or coordination

The complexion, also known as coordination, is the process by which cations attract molecules or anions that have a free electron pair. Coordination occurs between a central atom, anions or molecules with a heavy metal cation through a ligand. A "ligand atom" is a group of basic or nucleophilic atoms. Chelation is the process of generating a complex with multidentate ligands, which are bases containing more than one ligand atom **(Abdia & Kazemia, 2015)**. Electrostatic interaction between a polymer released by bacteria and a metallic ion chelating agent leads to complexation or coordination. Metals may be removed from the solution through complex formation on the cell surface after interacting with active groups on the cell wall, including carboxyl, amino, thiol, hydroxy, and hydroxyl carboxyl **(Sag & Kutsal, 2001)**. Electrostatic attraction exists between electron pairs in these chelating compounds **(Gahlout *et al***., **2021)**.

2.3. Physical adsorption

The movement of ions from a fluid to a solid state is the physical adsorption. The process, which is often fast and highly reversible, may take place at the surface due to non-specific attraction forces (such as Van der Waals forces) or due to coulombic attraction forces between charged solute species and the adsorbing phase **(Javanbakht *et al*., 2013)**. Since it is metabolically independent, this approach is particularly promising for treating enormous quantities of wastewater **(Gahlout *et al*., 2021)**.

2.4. Precipitation

During precipitation, metal ions attach to the biosorbent's surface functional groups and either remain unaffected or taken up by the microorganism. Through sorption-precipitation, metals may accumulate as organic or inorganic metal precipitates within cells or on cell walls **(AjayKumar *et al*., 2009)**. According to **Ahalya *et al*. (2003)**, the metabolic changes that take place in the cell following the metal's chemical contact with the cell surface may or may not have any bearing on the onset of precipitation **(Javanbakht *et al.*, 2013)**.

3. Factors affecting biosorption

Biomass and metals are only two of the many factors that play a role in biosorption processes; environmental factors also play a role **(Ghosh *et al*., 2016)**. The following are the primary elements that influence the biosorption process:

3.1. The pH values

The pH of a solution influences the kind of biomass binding sites and metal solubility, as well as metal solution chemistry, functional group activity in biomass and metallic competition in biosorption processes **(Deng & Wang, 2012)**. Metal biosorption has been demonstrated to be substantially pH-dependent in nearly every system examined, including bacteria, cyanobacteria, algae and fungi. Biosorption of metals such as copper, cadmium, nickel, cobalt and zinc is typically decreased at low pH levels owing to competition for binding sites between cations and protons **(Deng &Wang, 2012)**. Metal ion removal from solutions is minimal at pH levels below 2; when the pH rises from 3.0 to 5.0, metal absorption rises. The appropriate pH level is crucial for achieving maximal metal sorption, and as the pH value rises, so does this capacity **(Oyewole *et al*., 2018)**.

3.2. Temperature

The temperature has a substantial impact on the sorption process **(Farooq *et al*., 2010)**. Thermodynamic parameters are altered by temperature change, which affects sorption capacity **(Zeraatkar *et al*., 2016)**. The best temperature range for biosorption efficiency is between 20 and 35°C **(Aksu & Dönmez, 2001)**. High temperatures such as 50 degrees Celsius may occasionally improve biosorption, but they can also permanently destroy live microbes, reducing metal absorption **(Ahalya *et al*., 2003)**. Adsorption increases with decreasing temperature, which is generally due to exothermic absorption events.

3.3. Characteristics of the biomass

One important factor is the kind of biomass or waste used, whether it is applied as freely floating cells, immobilized cells or living biofilms. Physical treatments, viz. boiling, drying, autoclaving and mechanical disruption affect binding capacity, and chemical treatments such as alkali treatment often increase biosorption capability. This is especially essential in fungal systems where chitin is acetylated to form chitosan-glycan complexes with higher metal affinity **(Wang & Chen, 2009)**. Immobilized biofilms' surface area, as well as cells or particles may be impacted. As biomass content increases, sorption per unit weight decreases, which may affect biosorption efficiency **(Ahalya *et al*., 2003)**. Physical and chemical factors influence the metal affinity, permeability and surface charges of biosorbents. To increase the amount of metal absorption, the biomass may be manipulated using alkalis, acids, detergents and heat **(Wang & Chen, 2009)**.

3.4. Biomass concentration

The biomass concentration in the solution has a major impact on the specific uptake; as the biomass concentration decreases, the specific uptake rises. Biomass concentration grows, interfering with binding sites. The allegation is that the solution is deficient in metal concentration. As a result, when employing microbial biomass as a biosorbent, this issue must be considered. Metal ions' ability to reach binding sites is hampered by high biomass content **(Gadd, 2010)**.

3.5. Initial metal ion concentration

Biosorption is also affected by the initial metal ionic strength. Consequently, given the initial metal concentration, biomass will absorb a substantial amount of metal. The ideal percentage of metal removal may be attained at the beginning metal concentration, such that metal uptake increases as the starting concentration increases for a certain concentration of biomass **(Zouboulis & Matis, 2009)**.

3.6. Contact time

The quantity of total biosorption is determined by the contact time of the biosorbent. Biosorption rises with increasing contact time until it reaches the optimal contact time, at which point it stays practically constant. When all active sites are occupied, the biomass becomes saturated, resulting in an equilibrium **(Ibrahim *et al*., 2016)**.

4. Types of biological biosorbents

Various biomaterials as shown in Fig. (2) have a high biosorption capacity for all types of metal ions. Several types of biomasses have shown sufficient metal absorption capacity to warrant further investigation. Many biomaterials have been evaluated for absorptive properties and are classified as follows:

4.1. Plant products

Plant-based biosorbents are mostly agricultural byproducts, such as maize cob and husk **(Igwe *et al*., 2005)**, sunflower stalk **(Gang & Weixing, 1998)**, *Medicago sativa* (alfalfa) **(Dhankhar & Hooda, 2011)**, cassava waste **(Abia *et al*., 2003)**, wool, rice, exhausted coffee **(Dakiky *et al*., 2002)** and waste tea **(Ahluwalia & Goyal, 2005)**.

4.2. Algae as biosorbents

Algae are efficient and low-cost biosorbents of metal ions that are formed on the cell surface as a result of the ion exchange process. Algal biosorbents are freshwater and marine microalgae and macroalgae. Brown seaweeds, a kind of marine algae that comprises red, green and brown algae are effective biosorbents **(Davis *et al*., 2003)**. Brown sea algae have a high metal absorption capability for Cd, Ni and Pb through surface chemical groups, such as carboxyl, sulfonate, amino and sulfhydryl **(Mustapha & Halimoon, 2015)**. **Davis *et al.* (2003)** has discussed the capacity of seaweeds to absorb metals and discovered that *Sargassum* is one of the best seaweeds for heavy metal biosorption.

4.3. Bacteria as biosorbents

Bacteria have developed various effective detoxification methods for metal ions. They evolve resistance mechanisms primarily to ensure their survival in a polluted environment. Bacteria may bind metals due to anionic functional groups in their cell walls. Gram-negative bacteria have anionic functional groups due to the presence of peptidoglycan, phospholipids and lipopolysaccharides in their cell wall, and gram-negative bacteria containing peptidoglycan, teichoic acids and teichuronic acids; however, gram-positive bacteria do not **(Dhankhar & Hooda, 2011)**. Although large-scale separation, screening and harvesting of bacteria are difficult, it remains one of the most effective methods of pollution remediation **(Hrynkiewicz & Baum, 2014)**. *Bacillus, Streptomyces, Micrococcus, Pseudomonas, and Escherichia coli* are all powerful metal biosorbents **(Ansari *et al*., 2011)**.

4.4. Fungi as biosorbents

Fungi are a broad group of eukaryotic microorganisms that include species growing as unicellular yeasts and/or filamentous fungi with branching hyphae that produce spores and other reproductive features **(Mohmand *et al*., 2011; Carris *et al*., 2012)**. Since sorption is a surface reaction, it is possible to claim that the performance of the biosorbent is affected by the ionic state of the biomass. Consequently, the surface area and polarity of a biosorbent affect its biosorption capability **(Dhankhar & Hooda, 2011)**.

Fungi are regarded to be ideal candidates and decomposers for heavy metal biosorption **(Naveena & Latha, 2018)**. Potent metal biosorbents among fungi include genera such as *Rhizopus*, *Aspergillus*, *Streptoverticillum* and *Saccharomyces* **(Puranik & Paknikar, 1997; El-Bondkly & El-Gendy, 2022)**. Various studies have reported the ability of fungi for heavy metal biosorption. It has been reported that, the mycelium of the filamentous fungus *Phanerochaeta chrysosporium* may function as a biosorbent for cadmium, lead, and copper, while *Saccharomyces cerevisiae* can act as a biosorbent for zinc and cadmium removal **(Say *et al*., 2001; Chen & Wang, 2007; Talos *et al*., 2009)**. **Tigini *et al*. (2010)** reported that, *Cunninghamella elegans* is a promising biosorbent for the removal of heavy metals in textile wastewater. *Alternaria alternata* and *Penicillium aurantiogriseum* have also been identified as good biosorbents for cadmium and mercury removal **(Bahobil *et al*., 2017)**. Additionally, copper biosorption by *Ganoderma lucidum* and *Aspergillus niger* was postulated in the study of **Muraleedharan and Venkobachar (1990)**.

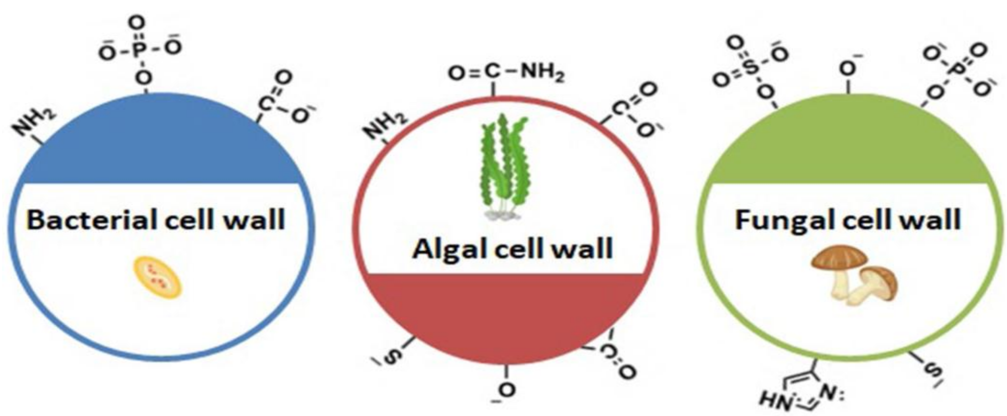

Fig. 2. Different biosorbent materials and their functional groups involved in biosorption **(after: Giese, 2020)**.

5. Advantages of fungi as biosorbent

Fungi have several benefits over other microorganisms such as bacteria. Due to the distinctive characteristics of the fungal cell wall surface that gather metals on their cell surface, fungal biomass has garnered a lot of interest as a biosorbent due to functional groups found on the cell surface such as carboxyl, amine hydroxyl, phosphate and sulfhydryl which are involved in heavy metal removal by different absorptive mechanisms as shown in Fig. (3) **(Kisielowska *et al*., 2010; Legorreta-Castañeda *et al*., 2022)**. They can endure variations in the food supply as well as other environmental conditions, including pH, aeration and temperature, apart from their capacity to bind metals **(Dhankhar & Hooda, 2011; Kanamarlapudi *et al*., 2018)**. Additionally, fungi represent an excellent metal sorbent and one of the industrial fermentation waste biomasses. As a consequence, it is feasible to produce and use fungal biomass on a large scale in industrial applications such as the bioremoval of metal ions from highly contaminated effluents using low-cost growing media **(Leitão *et al*., 2009; Dusengemungu *et al*., 2020; Taha *et al*., 2023)**. **Kanamarlapudi *et al*. (2018**) also discovered that the majority of fungal biosorbents are non-toxic, nontoxic and controlled.

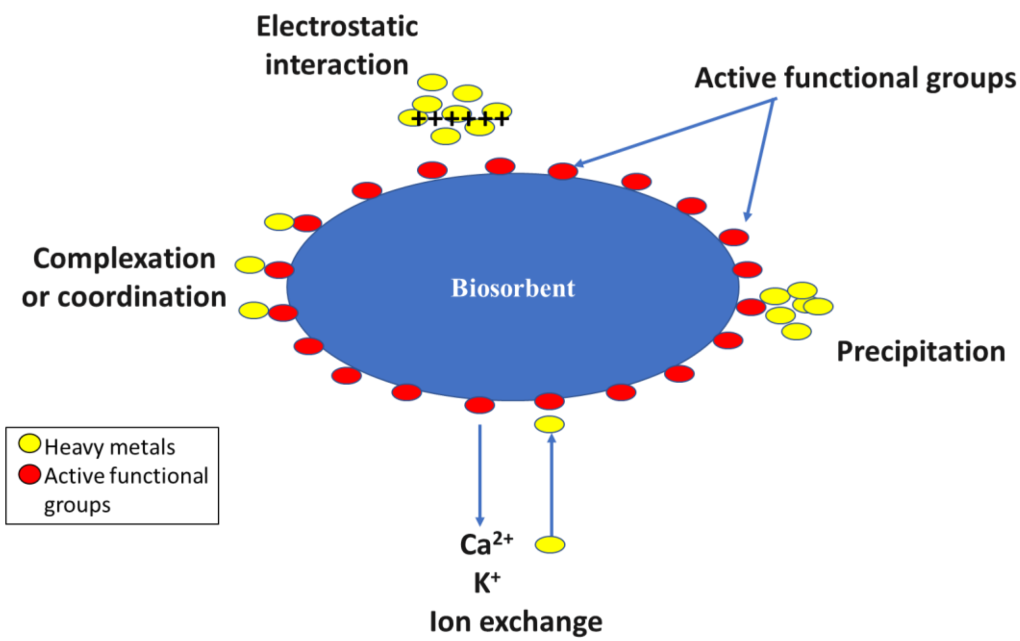

Fig. 3. Different biosorption mechanisms involved in heavy metals removal using fungi as biosorbent.

6. Fungal cell wall

The fungal cell wall has several favorable qualities that improve metal absorption and binding abilities. The fungal cell wall may account for up to 30% of the fungus's dry weight. The form and integrity of fungal cells are reliant on cell wall strength, which plays a range of critical functions when the fungus interacts with its surroundings such as sequestering metal ions **(Svecova *et al*., 2006)**.

The fungal cell wall is largely made up of polysaccharides, which make up about 90% of the cell wall. Many different proteins are heavily glycosylated; they are connected to polysaccharides in a variety of ways. These proteins are found in lower concentrations (3-20%), while lipids, pigments, and inorganic salts are significantly less prevalent, see Fig. (4). Polysaccharides, which are a diverse group of proteins that are often glycosylated, are anchored to the cell wall in a variety of ways, including lipids, proteins, pigments, polyphosphates, and inorganic ions **(Tan & Cheng, 2003).** Chitin is a common constituent of fungal cell walls. It is a polysaccharide that contains nitrogen and is made up of N-acetylglucosamine residues. The positively charged chitin or chitosan complex is often interleaved with negatively charged phosphates and glucuronic residues in cell walls. This contributes to the investigation of different heavy metal sequestration techniques **(Dusengemungu *et al*., 2020).**

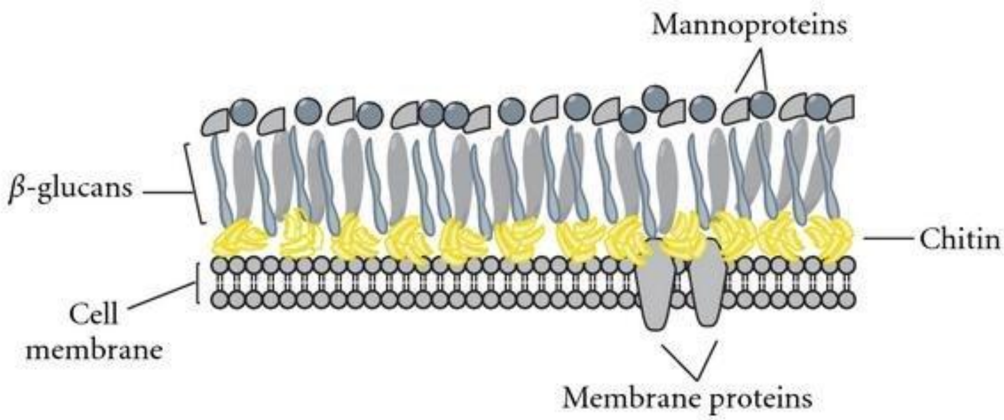

Fig. 4. Fungal cell wall composition **(after: Vega & Kalkum, 2012)**.

7. Metal biosorption by living and dead fungal biomass

Given the hazards of heavy metal toxicity, several studies on the removal of heavy metals from wastewater using both living and dead fungal biomass have been conducted, either via metal biosorption to biomass or around hyphae, have been conducted in the past two decades **(Ayele *et al*., 2021)**. The distinction must be made between active, metabolically mediated metal absorption by live biomass and passive metal sequestration by inactivated (dead) biomass. Metal absorption occurs independently of cellular metabolic activity and is characterized by metal ion surface binding to cell walls and extracellular substances, a process known as biosorption, or passive uptake as shown in Fig. (5). Metal absorption, on the other hand, is dependent on cell metabolism and is referred to as active uptake or bioaccumulation **(Abbas *et al*., 2014).**

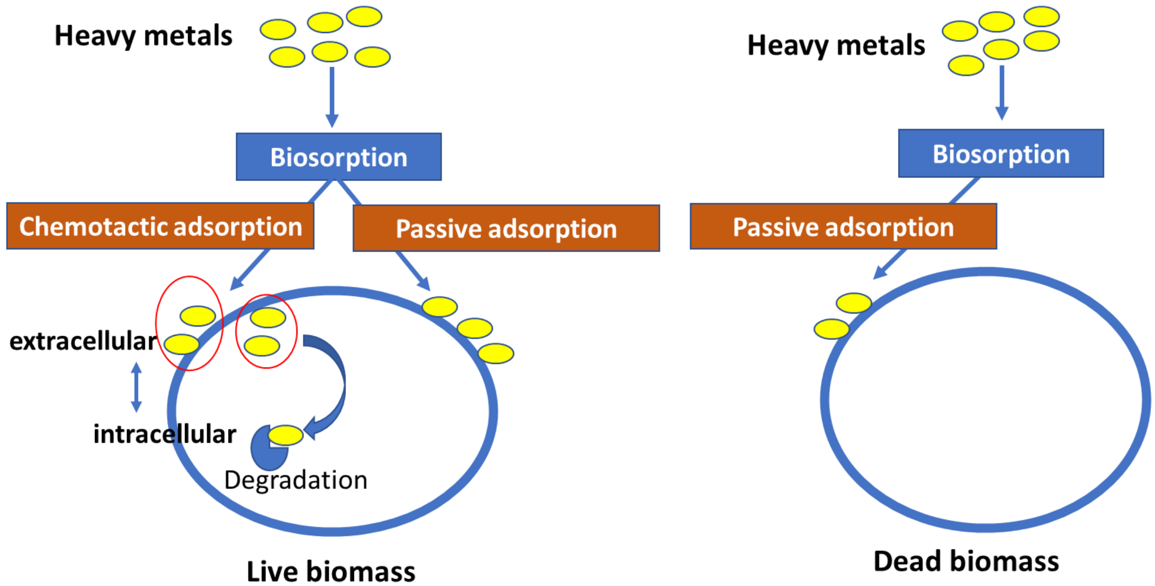

Fig. 5. Schematic diagram of metal biosorption by living and dead fungal biomass.

The terms "bioaccumulation" and "biosorption" seem to be gaining popularity. As a type of defense, actively metabolizing cells may aggressively resist metal ions, particularly the more dangerous ones. Consequently, biomass absorbs comparatively little metal. More metal ions may be gathered from the solution via the chemical binding sites of metabolically inactive cells (dead biomass) **(Naja & Volesky, 2011).**

Dead fungal biomass (biosorbent) is more effective at metal absorption than living fungal biomass and has been established as a suitable biosorbent **(Legorreta-Castañeda *et al*., 2020; Tayang & Songachan, 2021).** Dead biomass offers numerous benefits, including strong environmental resilience, higher toxicity tolerance, relatively quick regeneration, high sorbed metal recovery, easy numerical modelling of metal absorption reactors, and no requirement for a particular culture medium to keep it alive. Another advantage of employing inactive biomass is that it does not need nourishment or maintenance and may be kept for extended periods without affecting its function **(Javanbakht *et al*., 2013; Da Rocha Ferreira *et al*., 2019).**

Inactivated (dead) biomass is mostly derived from industrial sources because of different fermentation processes **(Abbas *et al*., 2014)**. Physical and chemical treatments may also be used to inactivate fungal cells for biosorption. Boiling, autoclaving, and freeze-drying are examples of physical procedures. One of the chemical techniques is the treatment of biomass with various organic and inorganic compounds such as dimethyl sulfoxide, laundry detergent, orthophosphoric acid, formaldehyde, glutaraldehyde, and NaOH **(Das *et al*., 2008)**.

CONCLUDING REMARKS AND FUTURE CONSIDERATIONS

Without treatment, industrial wastewater is harmful to the environment. Because heavy metal ions are toxic substances discharged into the water, they must be removed from mining and other industrial effluents. Because traditional removal processes are costly, it is vital to seek low-cost, ecologically friendly substitute solutions. According to the aforementioned literature analysis, biosorption is the most cost-effective, economical, and ecologically acceptable way of eliminating heavy metals from home and industrial wastewater. Several biological materials can be used as heavy metal biosorbents. Microbial biomass is one of the most cost-effective and feasible methods of eliminating heavy metals from environmental solutions. Fungal biomass is as good as or better than other microbial adsorbents for the removal and recovery of metals from wastewater due to its greater flexibility under prolonged exposure to contaminants, and it can be easily recovered and reused. Some fungi can survive and thrive in environments with high levels of hazardous metal ions. The removal of metals over a broad pH and temperature range is one of the most attractive aspects of the fungal biosorption process. Fungi may interact with heavy metals via a variety of chemical forces due to the huge number of polysaccharides, proteins, and large functional groups in the fungal cell wall. As a result, future studies should focus on discovering and analyzing the potential of fungi that can tolerate high levels of hazardous metals for use in biosorption studies, which might replace current metal-removal methods. Consequently, biosorption technology gains advantages over traditional approaches and grows in popularity.

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