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Comprehensive Evaluation of Mycoremediation using *Aspergillus* sp. Exopolysaccharide as A Sustainable Strategy

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Abstract

Heavy metals are naturally occurring and necessary for life; they can also accumulate in organisms and cause harm. The most prevalent heavy metals that can harm human health and cause poisoning are lead, mercury, arsenic, cadmium, chromium, copper, nickel and chromium. The biotechnological term "mycoremediation" refers to the efficient and economical deletion of hazardous contaminants from the environment using fungi. By using fungal biomass, heavy metals can be eliminated by biological processes as breakdown, absorption, accumulation, and conversion. Certain research has shown the effectiveness of numerous fungal species in eliminating heavy metals, such as mold (*Aspergillus; Rhizopus*), yeast (*Penicillium; Saccharomyces*), and mushrooms. However, filamentous fungal sp., such as *Aspergillus* sp., is the most adept at doing so from liquid substrates. The exopolysaccharide (EPS)-producing and bioremediation properties of *Aspergillus* sp. are known properties of the bioactive form. *Aspergillus* sp. EPSs is significant natural biopolymers that have potential to safely and efficiently detoxify the heavy metals from environment. They have a very promising future for the industrial sectors that produce heavy metals.

Keywords: Mycoremediation, Wastewater Treatment, Heavy Metals, *Aspergillus* sp., Exopolysaccharide, Eco-friendly, Cost Effective

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INTRODUCTION

High level of heavy metals induces genetic mutation in organisms that causes abnormalities and health problems.[1,2] Toxic metallic elements are poison for terrestrial and aquatic ecosystems which is a serious environmental concern. These toxic heavy metals have been detoxifying by various treatment techniques. Anthropogenic pollution that are caused by human activities like metallurgical industry, agricultural waste and sewage releases heavy metals that are hazardous metallic elements and affects food chain and disturb the ecosystem causes human health problems.[3] Human come into the contact with these heavy metals by consuming food, drinking water, breathing air and skin absorption that lead to negative health impact.[4] Conventional methods were used for the remediation of heavy metals.[5] An environmentally sustainable and economical approach for wastewater management is necessary to maintain the ecosystem as well as biodiversity. Microorganisms such as Fungi and Bacteria have potential to detoxify the heavy metals from the wastewater that could be a good alternative for treatment technique. Many fungal strains can eliminate heavy metals by bioaccumulation and biosorption process in a low cost and economical way. Fungi have biochemical capabilities that can break down organic molecules in the environment that reduces the risk of heavy metals. Fungal strains are perfect for mycoremediation because they have catabolic enzymes that form an extensive mycelia network that uses pollutant material for their growth.[6] *Aspergillus* sp. is a filamentous fungus found in saline water bodies. The bioactive form *Aspergillus* sp. is known to have exopolysaccharide (EPS) producing and bio-remediating effect. EPS is well-recognized to have significant effects on metal ion adsorption. Heavy metals can bond to EPS by micro-precipitation and proton exchange mechanism.[7] A summary of *Aspergillus* sp. EPS bioactivity is given in this review, along with recommendations for using mycoremediation for heavy metal biosorption, precipitation, biotransformation, and sequestration.

Principal genera of fungi for the Mycoremediation approach

Globally, about 69000 fungal strains have been found out of which selected fungal strains have been known for their uses in mycoremediation. Interspecific microbial competition is the cause of the biotic and abiotic factors and growth of fungal strains.[8]

Targeted fungal species for mycoremediation of heavy metals

The cell wall of fungal strains has metals binding polysaccharides and proteins. Metal removal efficiency is evaluated by selecting metal tolerant fungal strains.[9] Three fungal species, i.e. *Hymenogaster* spp., *Scleroderma citrinum*, and *Pisolithus tinctorius* have the highest potential to remove copper, zinc, aluminum and iron. Every fungal strain have specified tolerance activity against the selective heavy metal that depends of the type and the amount of heavy metals concentration.

Fungi have most significant bio mechanism such as bioaccumulation, bioadsorption, biosynthesis, biomineralization, bioreduction, bio-oxidation, extracellular and intracellular precipitation, external surface resorption towards the toxic metals tolerance and removal. The key factors influencing the mycoremediation of HMs amount of fungal biomass, pH, temperature and time duration between the contact of fungal strains and HMs. These variables change depending on the kind of HMs and respective potential of fungal species.[10] We have covered the appropriate fungal strain, their removal and tolerance mechanisms, and the variables influencing the elimination of heavy metals in this review.

Trichoderma harzianum has been identified against the selective metals such as lead, zinc, cadmium, nickel and copper by secreting organic acids which enhance the solubility of metals.[11]

Aspergillus terreus shows significant resistance to copper and can efficiently biosorb from water solution. Research indicates that *A. terreus* can eliminate as much as 68.25% of copper from water based solutions.[12]

Aspergillus niger is recognized for its capability to generate citric acid which can solubilize and chelate HMs such as Cd and Pd. It has been researched that *A. niger* can decrease Cd level as much as 87% in 14 days and also shows a strong tolerance against Pd and As in polluted water and soil.

Exopolysaccharides (EPS) from *Aspergillus* sp. as a natural acidic microbial compound contains uronic acid, pyruvate and organic acids that can easily binds with metal ions through interaction with negatively charged functional group to mitigate the metal toxicity. EPS have fiber like structure and have mycelia growth that can create an intermediate for active absorption of HMs and break contaminants to non-toxic products which is safe for environment.

Fungal sp. such as *Aspergillus transmontanensis, Cladosporium cladosporioides*, and *Geotrichum candidum* are effectively used for mycoremediation against heavy metals.[13] The EPS metal binding activities mostly depends on several factors such as pH, temperature, agitation, composition of media and biomass.[14] It has been already researched that the diffusion of HMs in EPS structure has been mathematically explained about chemical sorption activity. Figure 1 illustrates a few fungal species that have evolved a bioremediation mechanism to remove heavy metals that have been elevated by human activity[15] and have disrupted environmental compositions, endangering human health. The fungal strains listed in Table 1 possess the metabolic mechanisms necessary for Mycoremediation of the targeted heavy metals.

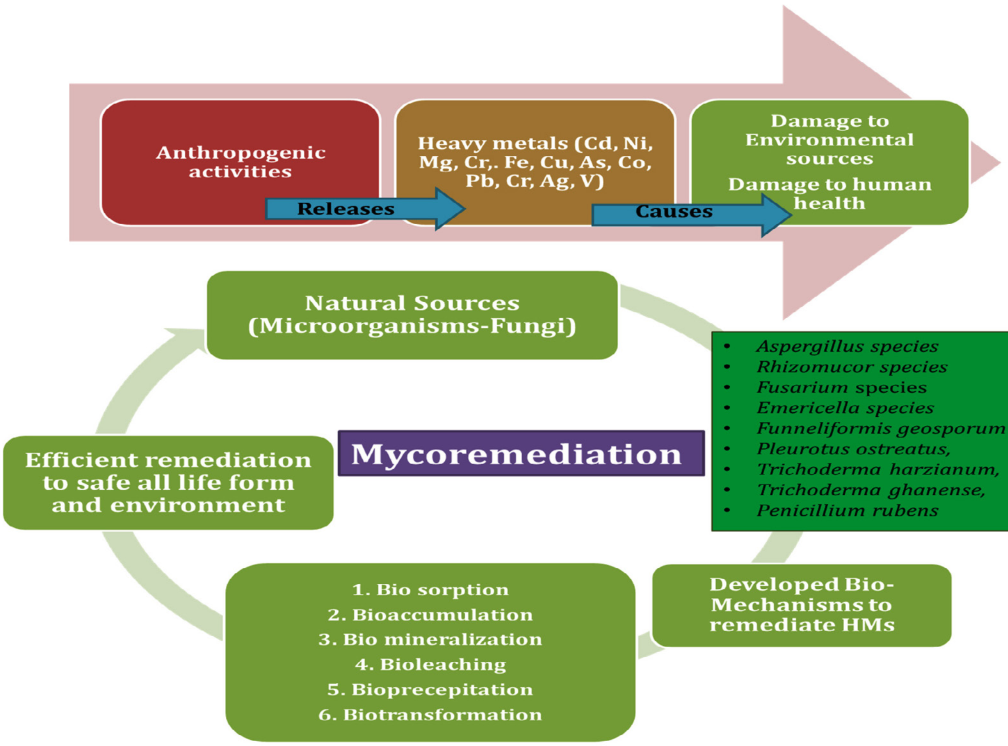

Figure 1. Specified fungal species for Mycoremediation of heavy metals

Table 1. Categorizing fungal strains based on their target metals

Fungal Strains	Selective heavy metals	Removal activity	Ref.
Aspergillus niger	Cr, Co, Cu	Biosorption	[16]
Aspergillus fumigates	Pb	Biosorption	[16]
Pencillium chrysogenum XJ-1	Cd	Biosorption	[16]
Corprinus comatus	Hg	Biosorption	[17]
Trichoderma brevicompactum QYCD-6	Cr, Pb, Cu, Cd	Bioaccumulation	[17]
Sterigmatomyces halophilus	Zn, Pb	Biosorption	[18]
Acremonium persicinum	Cu	Biosorption	[19]
Aspergillus flavus	Cu	Bioaccumulation	[20]
Saccharomyces cerevisiae	Cr	Bioaccumulation	[20]
Penicillium simplicissimum	Cu	Biosorption	[16]
Penicillium sp.	Co	Biosorption	[21]
Paecilomyces sp.	Co	Biosorption	[21]

Comparative analysis of *Aspergillus* sp. from other fungal sp.

The *Aspergillus* sp. surface cell wall is formed of chitin, proteins, polysaccharides polyphosphates and mineral ions that function as ligands to label and bind with metal ions and degrade them into less toxic compounds.[21] Table 2 summarized the specific studies conducted by different researchers to show how fungi can remove heavy metals from soil.

In 2017 bioremediation by bacteria associated with fungi which was the subject of study in China; for the behavior related to pollution by PAHs and heavy metals.[11]

Unique traits specific to *Aspergillus* sp.

Aspergillus sp. was first described by Spegazzini in 1896 and is strictly asexual.[22] *Aspergillus* sp. produces huge amounts of exopolysaccharides (EPS) that act as an adsorbing agent for metal recovery. More than 60% of all industrial enzymes derived from filamentous fungi are produced by *Aspergillus.* Because of their great secretion machinery capabilities, *Aspergillus* is a potentially useful host. *Aspergillus* sp. may be used as bioremediation of arsenic ions and can detect as biosensors also.[23] *Aspergillus penicillioides* has been identified to detoxify the cadmium and lead from the wastewater.[24] Figure 2 provides a complete overview of the various facets of *Aspergillus* sp. that can improve the use of Mycoremediation in the social sector.

Table 2. The studies listed below show that Aspergillus sp. is among the most favorable fungal sp. to use for heavy metal removal

Demonstrated *Aspergillus* sp.	Targeted Heavy Metals
Aspergillus niger	Pd and Phosphate.
Aspergillus terreus	Pb, Me, and Cd
Aspergillus niger	Zn and Cd
Aspergillus niger	Cd and Cu
Aspergillus sp.	Cu

Harnessing *Aspergillus* exopolysaccharides*:* A promising approach for heavy metal remediation

Fungal Exopolysaccharides (EPSs) are vital, safe, natural biopolymers that can be utilized as an environmentally friendly method of removing heavy metals from the environment because of their metal binding activity. The biosorption of EPS is cost-effective and safe because of non-chemical procedures. EPSs are potential conventional chemical polymers due to their high efficiency, nontoxic features, and biodegradability. Fungal extracellular polymers (EPSs) can consist of either pure sugars or sugars mixed with another unit, like protein and phosphate have sites to bind with metal ions. EPS consist of following functional groups in the composition as group of R-COOH, PO_4, $R\text{-}NH_,$ R-SH, Ar-OH and R-OH that provide a strong metal-ion sorption. EPS have the ability

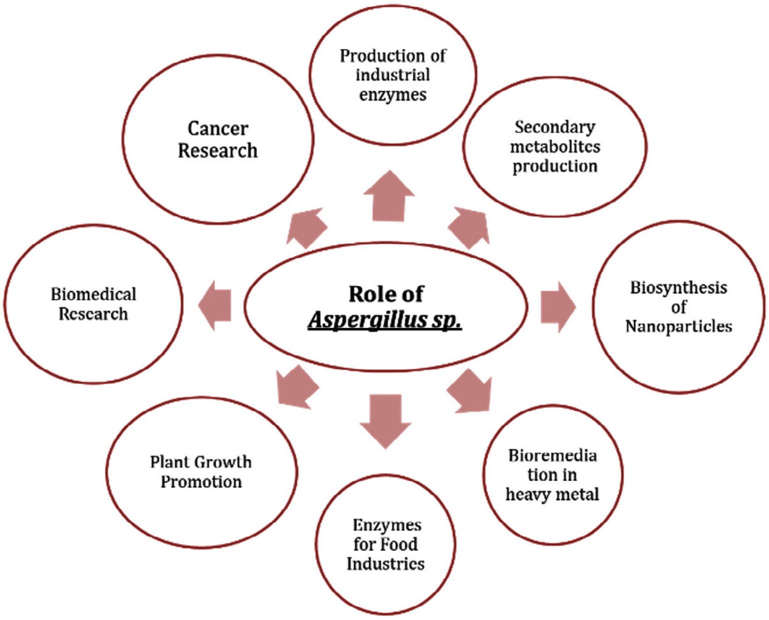

Figure 2. Beneficiary aspect of *Aspergillus* species in different sectors

Table 3. Identification of *Aspergillus Exopolysaccharide* composition and its metal remediation efficiency

Aspergillus sp.	EPS Composition	Targeted heavy metal with remediation efficiency	Ref.
Aspergillus alliaceus	Galactosamine, Galactos, Glucose, Acetate	Ferrous (68.1%), Zink (100%), Cobalt (71.4%)	[26,27]
Aspergillus niger	Glucose	Lead (88%), Cadmium (98%), Copper (84%)	[28]
Aspergillus parasiticus	Galactosamine, Galactose, Glucose, Acetate, Phosphate	Remediation of oil spills (80%)	[7,29]
Aspergillus fumigatus	Glucose	Copper (87%) Lead (43%)	[30]
Aspergillus versicolor	Glucopyranose, Mannopyranose, Glucan	Chromium VI (99.8%) Copper (30.5%)	[31]
Aspergillus ochraceus	Galactomannan	Copper (57%), Zink (57%), Chromium (38%)	[32]
Aspergillus penicillioides (F12)	Mannose, Galactose	Lead (92.4%), Cadmium (80%), Mercury (99.6%)	[33]

to adsorbed about 80% of lead and 30% of Cd.[25] Table 3 presented the content of EPS that was produced by various *Aspergillus* species' biocatalytic activity and used to remove heavy metals.

Aspergillus penicillioides F12 (MN210327) is the fungus strain that has demonstrated the highest level of heavy metal resistance activities against Hg(II), Pb(II) and Cd(II).[24] *Aspergillus terreus* K_1SF-Pb15 have shown metal removal efficiency up to 93% and *Aspergillus flavus* can remove 91% of Ni by biosorption activity.[24]

Rising Role of Fungal Exopolysaccharides in Emerging Applications

For the past 20 years, fungal EPSs acknowledged as high-value bio macromolecules. These products, which include botryosphaeran, scleroglucan, and pullulan have various uses in industrial sector, medical sector and food items among other things. Despite the great relevance of fungal extracellular polymers (EPSs), little is known about their biosynthesis to date, necessitating a thorough research look for fresh fungal strains that can produces unique EPSs.

In addition to providing a distinct source of marketable plant or seaweed polysaccharides, fungus-derived extracellular polymers (EPSs) have demonstrated a variety of novel and intriguing bio applications. In addition, compared to the manufacture of polysaccharides from plants or algae, the processing of fungal EPSs is simpler and a greater quantity can be produced in a shorter length of time.

Future prospects

Economic Significance

Aspergillus niger holds economic significance as fermentation organism that produces citric acid.

Environmental benefits

Aspergillus fumigatus is frequently grow in soil that compost piles to recycle carbon and nitrogen through the environment.

Agricultural benefits

Aspergillus niger appears to be a good bio-input for the creation of vegetable seedling because it increases the growth of all examined plants.

Industrial uses

Aspergillus niger used to produce food ingredients such as gluconic acid that control the acidity of wine and it is also a natural preservative. It is also used in the production of citric acid and various enzymes.

Aspergillus oryzae used to fermentation of soybeans, rice, potatoes and grains.

Medical benefits

A. terreus is widely utilized in industry to make xylanase and other essential organic acids and enzymes including itaconic acid and cis-aconitic acid. Additionally, it was the source of mevinolin, also known as lovastatin, a medication used to decrease serum cholesterol.

After the detailed review analysis of EPS metal ion binding it has been demonstrated that fungal EPS adsorb wide range of HMs it plays important role to resist the toxicity of HMs. A study by Paria *et al*. found that *A. Penicilloides* isolated from Subarnarekha River had a adsorption capacity to HMs.

Fungal EPS can be used for synthesis of bio-Nanosorbents that enhance the adsorption capacity of EPS metal ion interaction.

EPS can form a coating around the cell of microbes that finally produces the EPS for Mycoremediation of HMs in extreme geothermal conditions such as springs, saline lakes, and deep sea hydrothermal vents.

In Canada, *Aspergillus carbonarius* that was obtained from fermentation process found the removal capacity of Cu and Co at 11.6 mg/g whereas *Aspergillus flavus* extracted from soil get mitigate Cu and Co at 93.65 mg/g. So it is also shows that the application of EPS from different filamentous fungi have different removal capacity towards the HMs.

CONCLUSION

Heavy metals are major source of various neurological disorders in human body. For the bioremediation of heavy metals, numerous conventional techniques were employed, including adsorption, photocatalytic degradation, dialysis, coagulation, and filtering. Bacteria, fungi, and algae offer a viable solution for eliminating pollutants from wastewater, including heavy metals and dyes.

Not only is an eco-friendly alternative wastewater management program urgently needed for human health, but it is also necessary to preserve the survival of ecological diversity of the riverine ecosystem. Fungi belong to the detritus ecosystem of Earth. Using fungi to remove heavy metals from water, myco-remediation is an emerging in situ bioremediation technique that is both environmentally friendly and commercially successful.

Aspergillus sp. in its bioactive state is known to produce EPS and have a bioremediation impact. The pharmaceutical and food industries are the primary users of EPS, an exo-polymer.

In food industries, EPS can be used in food products as natural gelling agents that increase the texture of the product. EPS is a water binding agent that improves the moisture retention in food and increase the shelf life. Because of water binding activity property EPS is also used in cosmetic products that keep skin hydrated. As far as environmental concern EPS has explored as potential to biodegrade the plastic, some EPS contributes to form biofilms and biosorbent to remove pollutant from soil and water.

Toxic metallic compounds from wastewater have been removed for purification using a variety of biosorption processes. Fungal exopolysaccharides are vital, safe natural biopolymers that can be utilized as an environmentally friendly method of removing heavy metals from the environment.

Key Findings

Fungal EPS's biocompatibility, biodegradation and suitability for both people and environment make it important in versatile group of industries including agriculture, pharmacy, food and dairy. EPSs mediated metal reduction technique has an emerging green chemistry approaches.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

Both authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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DATA AVAILABILITY

All datasets generated or analyzed during this study are included in the manuscript.

ETHICS STATEMENT

Not applicable.

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