Applications of EPS in Environmental Bioremediations

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Abstract Micro-organisms (several fungi, bacteria, and algae) provide several biopolymers from renewable sources in which exopolysaccharides gain importance over other biopolymers. EPSs are useful for a vast variety of industrially important biomaterials. EPS accrues on the cell superficial of microbes and keeps safe against tough environmental conditions. In this book chapter, first, an introduction about EPS is given and then several current applications, potentials of exopolysaccharide which have been applied in bioremediation of heavy metals (HMs), colorant-containing residual, and toxic chemicals are deliberated.

Keywords Bioremediation · Exopolysaccharides (EPS) · Heavy metals · Environment · Biosorption · Dyes · Detoxification · Biofilm

Abbreviations

EPS	Exopolysaccharide
CPS	Capsular polysaccharide
HMs	Heavy metals
WHO	World Health Organization
PAHs	Polycyclic aromatic hydrocarbons

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1 Introduction

Exopolysaccharides (EPS) are complex biosynthetic polymers with microbial origins [10]. The polysaccharides from microbial origin are classified into two different forms (a) Capsular Polysaccharide (CPS) and (b) Exopolysaccharide (EPS) [88]. Depending on their location, EPS exhibit two typologies (a) Cell bound exopolysaccharides and (b) Released exopolysaccharide. Microbes secrete exopolysaccharide as carbohydrate polymers which form a layer on cell surface [103]. Exopolysaccharides (EPS) are organic macromolecule and water-soluble biopolymers attributed to various applications [118, 135]. Exopolysaccharides are divided into homopolysaccharides and heteropolysaccharides (HePS) (Molecular weight 10–30 kDa) [73]. Exopolysaccharides (EPSs) can be formed by prokaryotes (archaebacteria and eubacteria) and eukaryotes (Fungi, phytoplankton, and algae) have received more attention from scientists [145]. Different carbon sources, C/N ratio, pH, and temperature influence the composition and production of EPS [102]. EPSs are pondered as ubiquitous and plentiful bio-products [144].

The microbes are protected by exopolysaccharide against toxicity of heavy metal, drought, and salinity [26]. Exopolysaccharide has active and ionizable functional groups and substituents of non-carbohydrates which are responsible for the polymer's negative charge. Due to this property, ion exchange, complexation, and trapping, such as mechanisms, can bind different heavy metals to EPS and resulting in choice for the procedure of bioremediation [31].

Many microbial EPSs demonstrate the activity of metal ion biosorbent. Recently, heavy metal contamination has aroused in the environment and possess high risk to the health [81]. Heavy metal accumulation in environment due to manufacturing of electrical machinery, metal mining operations, tannery factories, municipal sewage disposal pesticides, and chemical fertilizers [119]. Biomineralization, biosorption, phytostabilization, mycoremediation, hyperaccumulation, rhizoremediation, cyano-remediation, dendroremediation, biostimulation, and genoremediation are among recent biotechnological tactics to bioremediation [87].

2 Environment Pollutants

Heavy metals are naturally occurring elements with relatively high density compared to H_2O and causes serious environmental issues [142]. Rapid industrialization is the main reason for the exposure of toxic heavy metals from soil colloids and water supplies. Heavy metal is persistent inorganic contaminants that are toxic to plants, animals, and humans due to food chain accumulation [152]. Heavy metal contamination is caused by waste from factories, and refineries, as well as indirectly by pollutants into the water stream [39]. Lead (Pb), copper (Cu), arsenic (As), mercury (Hg), cadmium (Cd) are considered to be the extreme common pollution causing heavy metals [61, 89].

Fig. 1 Heavy metal contamination on earth

Textile industry is mainly accountable for the water pollution that releases untreated sewage water into river, water bodies and poses risks to environment [65]. Dyes (synthetic) are broadly used in textile, paper, pulp industries, color photography, chemical, and cosmetics industries (Fig. 1) [12].

3 Sustainable Bioremediation of Toxic Heavy Metals

The application of EPS biopolymer started in 1960s, and their marketable uses have increased considerably since then [115]. Microbes can absorb and remove pollutants from the atmosphere through different pathways [33]. Initially, EPS research focused primarily on the treatment and elimination of toxic heavy metal from manufacturing as well as domestic wastewater [112]. The bioremediation of hazardous metals by microbes can be helpful in comparison of traditional physico-chemical procedures [95]. Conventional remediation approaches are either costly or produce dangerous by-products that adversely affect the atmosphere [49]. Due to physical, structural, and chemical diversity, scientists show the research curiosity in microorganisms' exopolysaccharides, and their uses in environmental bioremediation have increased [9, 72]. The bioremediation mechanism efficiently minimizes the hazard of contamination by removal of toxic metals from soil and groundwater [136].

In recent times, use of biomass (bacteria both living and dead conditions) for bioremediation has arisen as safe, low waste production, environment-friendly, cost-effective, low energy demand, and self-sustainability alternative for HMs removal (Fig. 2) [45]. Biosorption from microbes is also an innovative procedure for the management of waste [67]. The innovative exopolysaccharide might be used as novel source for heavy metals removal in various sectors. Adsorption of various HMs by bacterial EPS (*Bacillus firmus, Pseudomonas pachastrellae* KMS2-2, *Bacillus cereus* KMS3-1) have been reported [71, 126]. EPS-M816 (extracellular polysaccharide) obtained from *Mesorhizobium loti* (formerly known as *Rhizobium loti*) on glycerol-based media and is used as emulsifying agent [32].

EPS production is allied with biofilm processing which is important for biosorption and bio-mineralization of metal ion [41]. In 2020, Rusinova-Videva et al. [123] reported the proficiency of *Cryptococcus laurentii* (AL65) for heavy metal biosorption. Exopolysaccharide be able to chemically changed via acetylation, methylation, carboxymethylation, and sulphonation, that amends natural activities of EPS, thus refining the applicability of the polymer [104]. In harsh conditions such as geothermal springs, saline lakes, and deep-sea hydrothermal vents, EPSs often make a coating around the microbe cell [101]. Hydrothermal deep-sea vents may be a virtuous source of novel exopolysaccharide [47]. Various microorganisms are also reported for EPS production and used for environmental bioremediation (Table 1).

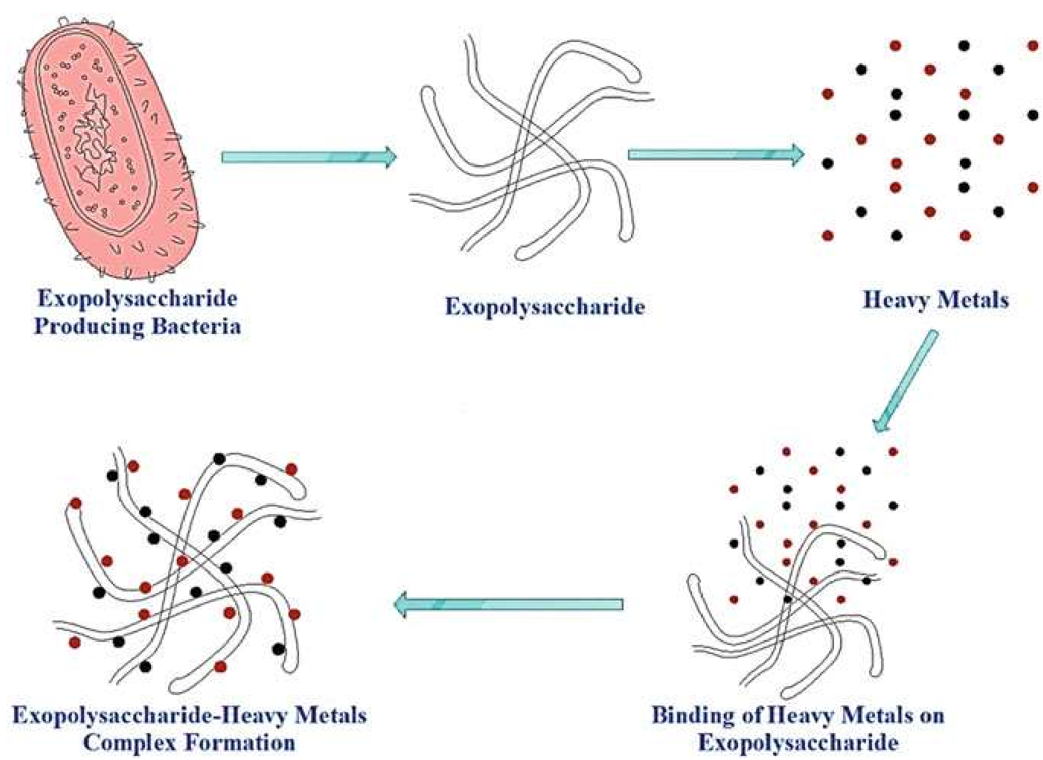

Fig. 2 Mechanism of exopolysaccharide interaction with heavy metals

Table 1 Production of EPS from microbes and their applications

EPS producing microbes	Applications	References
Bacillus licheniformis MS3	Excellent emulsifying properties against hydrocarbons and oils	[10]
Sphingomonas sp. MKIV	Ionic liquids (ILs) bioremediation	[70]
Klebsiella oxytoca strain DSM 29614	As(III) and As(V) removal from contaminated water	[16]
Bacillus strain TCL	Bioremediation of contaminated leather industry effluent and chromium reduction	[11]
Pantoea agglomerans	Metal bioremediation	[91]
Bacillus sonorensis MJM60135	Emulsification of toluene and bioremediation of hydrocarbons	[110]
Escherichia coli ATCC 25922 (ECBK) and *Staphylococcus epidermidis* RP62A (SEBK)	Removal of Cr(VI) and Zn(II)	[116]
Sphingomonas pseudosanguinis and *Sphingomonas yabuuchiae*	Bioremediation of biodiesel-derived waste glycerol (WG)	[117]
Bacillus cereus VK1	Bioremediation of Hg^{2+} polluted eco-systems	[64]
Alteromonas sp. JL2810	Removal of heavy metals Cu^{2+}, Ni^{2+}, and Cr^{6+}	[153]
Leptothrix cholodnii SP-6SL	Metal adsorption	[100]
Achromobacter xylosoxidans strain TERI L1	Produce EPS bioflocculant	[138]
Arthrobacter sp. SUK 1205	Bioremediation of environmental Cr pollution	[36]
Shewanella oneidensis	Adsorption of Cd(II)	[151]
Pseudomonas sp. Lk9	Removal of heavy metal ions (Cd^{2+} and Cu^{2+})	[84]
Synechocystis sp. BASO671	Metal removal	[108]
Arthrobacter ps-5	Biosorption capacity on metal ions (Cu^{2+}, Pb^{2+} and Cr^{6+})	[134]
Azotobacter chroococcum XU1	Biosorption of Pb and Hg	[119]
Paracoccus sp., *Alteromonas* sp., *Vibrio* sp., One unidentified	Cu(II) and Ag(I) bioremediation	[35]
Enterobacter cloacae strain P2B	Bioremediation of Lead	[98]
Rhizobium miluonense CC-B-L1, *Burkholderia seminalis* CC-IDD2w and *Ensifer adhaerens* CC-GSB4	Diesel oil emulsification	[56]
Pseudoalteromonas sp. A28 *Vibrio proteolyticus*	Biodegradation of a Bioemulsificant Exopolysaccharide (EPS_{2003})	[15]
Aspergillus fumigatus	Sorption efficiency copper and lead	[150]
Gordonia polyisoprenivorans CCT 7137	Removal of contaminating oil	[43]

(continued)

Table 1 (continued)

EPS producing microbes	Applications	References
Synechocystis sp.	Cr(VI) removal	[107]
Penicillium simplicissimum	Cd(II), Zn(II), and Pb(II) biosorption	[40]
Gordonia alkanivorans CC-JG39	Diesel biodegradation	[139]
Psuedoalteromonas sp. strain TG12	Emulsifying activity against oil substrate and metal binding activity	[51]
Paenibacillus jamilae	Production of a metal-binding EPS for adsorption	[93]
Pestalotiopsis sp. KCTC 8637	Wastewater treatment	[92]
Chryseomonas luteola TEM05	Cadmium and cobalt ions adsorption	[106]
Pseudomonas putida CZ1	Copper(II) and Zinc(II) biosorption	[21]
Bacillus subtilis	Adsorption of neutral Hg(II)	[30]
Cyanothece CE 4 and *Cyanospira capsulata*	Remove Cu^{2+} from aqueous solution	[34]

Bioremediation of Lead (Pb): Lead released from processing factories, recycling plants, and automotive industries [63]. In the environment, lead is a non-bioessential, hazardous, and pervasive metal that affects all living beings [97, 143]. The adsorption of lead by EPS has been investigated in several studies. Concordio-Reis et al. [24] reported that polysaccharide FucoPol (fucose-containing exopolysaccharide) secreted by *Enterobacter* A47 have good potential as biodegradable and safe biosorbent for the management of Pb^{2+} contaminated water. *Paenibacillus jamilae*'s EPS adsorbs Pb, Cd, Co, Ni, Zn, and Cu. The EPS showed good interaction with Pb with binding capacity of 303.03 mg/g [94].

Bioremediation of Cadmium (Cd): Cadmium is a persuasive pollutant and environmental contaminant that can unfavorably affect anthropoid health [20]. At extremely low concentrations of around 0.001–0.1 mg/L, this heavy metal is dangerous to individuals [140]. One of the greatest tactics of microbes to combat against toxicity of HMs is the exopolysaccharide (EPS) production [90]. Bacterial exopolysaccharide from *Arthrobacter viscosus* can significantly increase biosorption of Cd [132]. *Halomonas* sp., is an exopolysaccharide-producing halophilic bacterium which uptake more than 50% cadmium and might be used for bioremediation of Cd in polluted saline soils [7].

Bioremediation of Arsenic (As): According to World Health Organization (WHO), the concentration of As in drinking water is 10 µg/L [146]. As is toxic metalloid released into the atmosphere by natural and anthropogenic activities. The major activities include i.e., mining, weathering, combustion of fossil fuels and volcanic happenings Arsenic causes several severe diseases in human and animals [14, 82, 131, 141]. The polyanionic EPS produced biofilm is pondered as admirable biosorbent material for the bioremediation of arsenic and other metals [124].

Halomonas sp. Exo1 was proficient producer of exopolysaccharide and bio-remediate arsenic. Apart from As, EPS has also role in biosorption of Cr, Hg, and Mn [96].

Bioremediation of Mercury (Hg): Mercury is significantly dispersed in environment and found in the water, soil, and air. This metal is being highly poisonous to human beings, animals, and plants at low concentrations, so its remediation is incredibly essential [137]. Metal mining, fossil combustion, acetaldehyde industries, chloralkali plants, amalgamation industries, and paint industries are primary source of mercury pollution [80]. Mercury toxicity depends primarily on the speciation of Hg [4]. Mercury enters the environment mainly as Hg^{2+} from industrial sources [121]. Exopolysaccharide shows substantial role in mercury detoxification by heavy metal bioremediation method. *Vibrio fluvialis* was evaluated at four separate concentrations (100, 150, 200, and 250 μg/ml) for Hg removal potential and effective bioremediation was observed at 250 μg/ml (60% Hg removal) [129]. *Bacillus thuringiensis* strain RGN1.2 is used for bioremediation of mercury pollution and remove 96.72%, 90.67%, and 90.10% of mercury at 10, 25, and 50 ppm, respectively [114].

Bioremediation of Chromium (Cr): Chromium is an imperative metal for living beings, but it is also harmful at extremely low concentrations due to its high toxicity, and carcinogenicity [22]. Industrial development, such as electroplating, metallurgy, dyeing, paper, and pulp making, and tanning, leads chromium contamination in the environment [53]. *Enterobacter cloaceae* produced exopolysaccharide with Cr (VI) at 25, 50, and 100 ppm, and approx. 60–70% Cr was accumulated by this bacterium [58]. Hexavalent chromium [Cr(VI)] is a possible mutagen and carcinogen, which is introduced in the atmosphere by the anthropogenic activities [27]. In India, around Sukinda valley, pollution of hexavalent chromium was due to Chrome mining activity. Exopolysaccharide from *Enterobacter cloacae* SUKCr1D has been able to minimize Cr(VI) concentration by 31.7% at 10 mg/l [52].

Apart from the bacterial polysaccharide, some fungal EPS also produced by *Aspergillus terreus, Aspergillus niger, Lentinula edodes, Fusarium solani, Fusarium oxyporum* species [19, 48, 133]. Scientists have research focus on fungal polysaccharides in several biotechnological fields [105]. Basidiomycota and Ascomycota fungal strains, and yeast species are recognized for their capability to produce EPSs under different cultural environments [6]. The significant biological properties of exopolysaccharides produced by fungi were recognized only 20 years ago [147]. Since several studies have shown various applications in environmental bioremediation, fungal exopolysaccharide has also gained significance from last few decades [113, 120]. Mushrooms are known to be good producer of EPS and have applications in various bio-productive fields as well as play part in bioremediation of various heavy metals [23, 111]. Exopolysaccharides produced from fungi have also antioxidants, anti-cholesterol, and anti-cancer properties [85]. Algal extracellular polymeric substances (EPSs) are also used as bioflocculant in the bioremediation process [74]. The EPS from *Anabaena spiroides* (cynobacterium) was efficient in the binding capacity of Mn(II), Pb(II), Hg(II), and Cu(II) [42].

4 Applications of EPS in Bioremediation of Colorant-Containing Residual from Textile Effluent

On earth, water is the most vital resource for life. Textile industries are imperative for economic development for nations but it not only consumes enormous quantities of water but also pollute water bodies via discharge of dye effluents and are responsible for major environmental pollution [19, 76, 149]. Bioremediation is a biological process for the elimination of colorant-containing residual by microorganisms (Table 2) [109]. Azo dyes i.e., Methyl orange (MO) and Congo red (CR) are degraded by exopolysaccharide-stabilized silver nanoparticles (AgNPs) produced by *Leuconostoc lactis* KC117496. EPS-AgNPs are synthesized through reduction of Ag^+ into Ag^o from $AgNO_3$ which are effective, inexpensive, and environmentally safe candidate for biodegradation of hazardous textile dyes [130]. Li et al. [79] reported first time EPS-605 (a non-glucan EPS) that showed greatest biosorption capability for Pb^{2+}, Cu^{2+}, Cd^{2+}, and methylene blue. Halophilic *Exiguobacterium* sp. VK1 (indigenous salt pan isolate) removed carcinogenic Malachite Green with biosorption capacity of 79.1% at pH 6.0 and 40 °C temperature [65].

Pseudomonas aeruginosa and *Ochrobactrum* sp. produced EPS 456.4 mg l^{-1} and 404.6 mg l^{-1} after incubation for 48 h (40 °C) and 72 h (30 °C) respectively. EPS produced by *P. aeruginosa* and *Ochrobactrum* sp. removed Remazol Blue (RB) with a maximum yield of 12.5% and 89.4% respectively [68]. Novel hetero-exopolysaccharide-R040 (EPS-R040) prepared from *Lactobacillus plantarum* used as adsorbent for the removal of Methylene Blue [77].

5 EPS in Bioremediation of Oil Spills and Petroleum-Contaminated Sites

With the continual population growth, industrialization, and dependence on petroleum goods, environment pollution has increased globally. Petroleum oil-contaminated environment is not safe for both human and animals [5, 69, 148]. Oil biodegradation was measured respirometrically and based on variations in the composition of oil [122]. Ibrahim et al. [57] isolated halophilic bacterium *Halobacillus* sp. strain EG1HP4QL from Lake Qarun, Fayoum Province, Egypt that produced EPS and showed bioremediation prospective to consume crude oil (35.3%) as the solitary carbon source.

Vibrio harveyi (VB23) exopolysaccharide is hetero-polysaccharide and has strong activity of emulsification [13]. EPS produced by *Enterobacter cloaceae* (EPS 71a) emulsified benzene, hexane, kerosene, xylene, paraffin oil [59]. EPS produced from species of *Halomonas* have amphiphilic properties and contributed to the ultimate oil removal and oil aggregate formation [50]. *Sporosacina halophila* produced EPS, biosurfactant, and laccase for toluene biodegradation. It may also use for bioremediation of hydrocarbon adulterated sea and waste-water [1].

Table 2 Bioremediation of various toxic dyes by using exopolysaccharides

Name of dye	EPS producing microorganism	Application	References
Acridine orange and crystal violet	*Serratia* sp. ISTD04	Removal of dye	[75]
Azo dyes	*Rhizopus arrhizus*	Removal of azo dyes from aqueous dye solution	[127]
Methylene blue	*Rhodotorula mucilaginosa* strain UANL-001L	Methylene blue (MB) adsorption using EPS	[44]
Reactive dyes	*A. niger*, *A. oryzae* and *R. arrhizus*	Biosorption of textile dyes	[128]
Amaranth dye	*Bacillus* sp. AK1, *Lysinibacillus* sp. AK2, and *Kerstersia* sp. VKY1 strains biofilm	Decolorization of Amaranth dye	[8]
Congo red and indigo carmine	*Dietzia* sp. PD1	Biosorption of acid dye from aqueous solution	[125]
Remazol blue (RB)	*Pseudomonas aeruginosa* and *Ochrobactrum* sp.	Dye removal	[68]
Malachite green (MG)	*Pseudomonas aeruginosa* NCIM 2074	Decolorization process	[66]
Acid black 172	*Pseudomonas* sp. strain DY1	Biosorption of dyes from wastewater	[38]
Methylene blue	*Lactobacillus plantarum* JNULCC001	Excellent biosorption ability toward methylene blue	[78]
Reactive dyes	*Aeromonas* sp., *Pseudomonas luteola*, *Escherichia coli*, *Bacillus subtilis* and *Staphylococcus aureus*	Adsorption of reactive dyes	[54]

6 EPS in Bioremediation of Toxic Chemicals (PCBs, PAHs, CP)

Polycyclic Aromatic Hydrocarbons (PAHs) are environmental pollutants emitted from coal mining, power plants, and chemical industries. It is well-thought-out as a highly toxic pollutant because of their toxic, and carcinogenic properties [2, 46]. Exopolysaccharide (EPS) is produced by several species of rhizobia (soil bacterium) and play roles as bioemulsifier with possible applications for hydrocarbon degradation. *Rhizobium tropici* is member of the Rhizobiaceae and produced exopolysaccharides [17, 18]. *Rhizobium* sp. produced exopolysaccharide that showed flocculating and metal sorption properties [28]. Exopolysaccharide (MW $\sim 2 \times 10(5)$ Da) produced from *Klebsiella* sp. PB12 showed emulsifying activity with toluene, *n*-hexadecane, olive oil, and kerosene of 66.6%, 65%, 63.3% and 50% respectively [86]. Kachlany et al. [62] observed that hydrocarbon-degrading bacterium (*Pseudomonas putida* G7) produced EPS and show great metal binding capacity. EPSs of *Aspergillus niger* and *Zoogloea* sp. degrade more than 80% pyrene (polycyclic aromatic hydrocarbon) in polluted soils [60]. *Paenibacillus jamila*'s EPS contributes in bioremediation of wastewater generated during olive oil process [3]. Naseem and Bano [99] observed that the soil moisture, plant biomass was improved from *Alcaligenes faecalis* (AF3), *Proteus penneri* (Pp1), and *P. aeruginosa* (Pa2) (EPS producing bacteria) by seed bacterization of maize. *Enterobacter cloacae* strain TU (Gram-negative rod-shaped bacterium) release extracellular polymeric substance and exhibiting bio-emulsifying activity. The bacterium also utilizes alkanes and polycyclic aromatic hydrocarbons (PAHs) [55].

7 Concluding Remarks

Microbial exopolysaccharides are vastly diverse, sustainable, and environmentally safe alternatives in various applications especially in bioremediation. EPS producing microbes remediate the colorant residues, toxic chemicals, and hazardous heavy metals from environmental effluents. Biofilm-mediated remediation has been establishing as safer alternative to bioremediation. EPSs received significant research consideration from scientists in recent years because of their biodegradability and biocompatibility. According to existing information on exopolysaccharides, these biopolymers can cover a broad array of research in greener technologies.

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Conflict of Interest The authors declare that there is no conflict of interest.

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