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Microbial surface active compounds (SACs) for betterment of environment

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ABSTARCT

This review discusses the usage of Microbial surface active compounds (SACs) in reducing the negative impact imposed by water insoluble pollutants on environment. Microbial SACs either emulsify or increase surface area of insoluble pollutants such as polycyclic aromatic hydrocarbons (PAHs) and make them bioavailable for microbial degradation. Microbial enhanced oil recovery method also uses SACs making this method environment friendly and cost effective. Biosurfactants also detoxify and remove heavy metals and chlorinated compounds form water by making them bioavailable for degradation. Biosurfactants with less toxicity and lesser carbon footprint can be used in detergents.

Key words: *Microbial surface active compounds (SACs), Polycyclic aromatic hydrocarbons (PAHs), Biosurfactants, Bioemulsifiers, Bioremediation, Microbial enhanced oil recovery (MEOR).*

Introduction

Many anthropogenic activities including various industrial processes, agricultural processes, mining related activities, and activities of petroleum industries, etc. are responsible to add various water insoluble pollutants in environment. These pollutants such as heavy metals, polycyclic aromatic hydrocarbons, chlorinated compounds, chemical surfactants, etc. are imposing toxic effects on marine as well as terrestrial ecosystems. Various microorganisms are capable of degrading or detoxifying these pollutants such as microorganisms including fungi of the genera *Penicillium*, *Aspergillus*, *Candida*, etc., and bacteria from the *Actinobacteria*, *Alphaproteobacteria* and *Gammaproteobacteria* classes (Perdigão *et al.*, 2021).

As these pollutants are water insoluble, they are not bioavailable for microorganisms degrading them. The availability of these pollutants increases by adding surface active compounds (SACs) of biological origin. Microorganisms are well studied for their abilities of producing surface active compounds (SACs), such as high molecular weight bioemulsifiers and low molecular weight biosurfactants. These act at the crossing point by lowering the interfacial tension or by firmly binding to the surfaces (Ron and Rosenberg, 2002). Various microorganisms have been recorded for their ability to produce SACs. Rhamnolipids by *Pseudomonads*, glucose lipids by *Alcanivorax borkumensis*, emulsan by *Acinetobacter* RAG-1, a polysaccharide-protein complex by *Acinetobacter calcoaceticus*, alasan by *Acinetobacter radioresistens*, etc. are some examples of SACs produced by bacteria (Rosenberg and Ron, 1999).

Bioremediation of oil spills

Total petroleum hydrocarbon (TPH) in liquid form disposed into the terrestrial or marine ecosystem because of human actions is understood as an oil spill.

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Disposal of liquid petroleum hydrocarbons in marine water occurs because of various anthropogenic activities such as oil exploration, oil production, hull failure/collision/fire/explosion leading to accidents of oil tankers, routine ship operations, etc. More than 200 various oil spill incidences have been recorded in last 50 years, of which 140 were major. These major spills were responsible for releasing in excess of 7 million tons hydrocarbon oil into environment. Along with these anthropogenic oil spills, millions of tons of TPH are added to marine environment due to the natural seepage every year (Sayed *et al.*, 2021). Exxon Valdez spill befallen in Alaska on 24th March, 1989 and Deepwater Horizon spill happened in Gulf of Mexico on 20th April, 2010 are the two famous spills got well publicized. Volatile organic compounds (VOCs) as well as Polycyclic aromatic hydrocarbons (PAHs) are two core components of crude oil. After an oil spill, VOCs quickly dissipate in the environment and show acute toxicity to nearby inhabitants. PAHs remain in the environment for many years causing harm for many years to inhabitants after oil spill. These components of crude oil not only harm physically but also biochemically to the inhabitants affected by oil spill (Perdigão *et al.*, 2021). Remediation of these oil spills can be done in several ways, either individually or in combination. Mechanical methods are used for inhibition of oil from spreading, ex. booms, skimmers, sorbents, etc., thermal methods include *in situ* burning is a rapid oil removal method, chemical methods such as dispersion are used to control the spreading of oil, natural methods include bioremediation, which is nothing but the degradation of oil happening naturally with the help of microorganisms (Pete *et al.*, 2021). Bioremediation can be done with two different approaches; 1) bioaugmentation: the supplementation of oil degrading microorganisms at the oil spill site, 2) biostimulation: the supplementation of nutrients and growth limiting factors to boost the growth of native oil degrading microbes (Sayed *et al.*, 2021). Bioremediation is nothing but the speeding up of natural biodegradation process. The primary step of PAHs degradation requires direct contact between microbe and hydrocarbon substrate for catalytic activity of membrane associated oxygenase to be performed. As PAHs have low water solubility, surface active compounds (SACs) either emulsify them or increase their surface area to make them bioavailable (Ron and Rosenberg, 2002). Acceleration of biodegradation process can be done with addition of SACs or microorganisms producing SACs at the oil contaminated site. SACs help enhancing natural degradation process as they either make water insoluble PAHs bioavailable or increase their surface area. Various examples of using SACs in bioremediation of oil are known, some of which include the use of alasan produced by *Acinetobacter* and rhamnolipid produced by *Pseudomonas aeruginosa*in enhanced degradation of various PAHs (Ron and Rosenberg, 2002). Ron and Rosenberg (2002), also studied the *Acinetobacter radioresistens* known for production of alasan and for enhanced biodegradation of oil when cocultured.

Bioremediation of oil contaminated soil

Contamination of terrestrial ecosystems with petroleum and its derivatives is originated anthropogenically. Exploration of oil, transportation, refining, storage, etc., are some of the activities of petroleum industries responsible for contamination of soil with hydrocarbons (Stepanova *et al.*, 2022). These contaminants are mixture of various petroleum derivatives such as asphaltenes, alkanes, aromatic compounds, cycloalkanes, resins, etc., which have a very high toxic, mutagenic, carcinogenic impact on ecosystems. Contamination of soil with hydrocarbons changes its chemical and physical properties, which reduces soil's arable value as well as fertility (Ambust *et al.*, 2021; Sathe *et al.*, 2012). There are various strategies that have been devised for remediation of oil contaminated soil, such as physical methods: vapor removal, floatation, use of ultrasonic waves, electro kinetics remediation, biochar adsorption, thermal desorption, etc., chemical methods: plasma oxidation, photocatalytic degradation, etc., and biological methods: phytoremediation, bioremediation. Among all these methods, bioremediation is utmost cost-effective and eco-friendly (Stepanova *et al.*, 2022). Bioremediation of soil is a process used to degrade, detoxify, mineralize or transform hydrocarbon pollutants by using living organisms or their products (Yuniati, 2018). Several microorganisms belonging to different groups present ubiquitously have shown the ability to degrade petroleum hydrocarbons. Some of them are *Acremonium* sp., *Aspergillus fumigatus*, *Pseudomonas fluorescens*, *Aspergillus* spp., *Pseudomonas aeruginosa*, *Cephalosporium roseum*, *Mycobacterium smegmatis*, *Cladosporium cladosporoides*, *Nocardia petroleophilia*, *Fusarium* sp., *Desulfovibriode sulfuricans*, *Fusarium moniliforme*, *Candida tropicalis*, *Corynebacterium glutamicum*, *Rhodotorula* sp., *Bacillus* sp., *Torulopsis colliculosa*, *Arthrobacter simplex*, *Y arrowia tropicalis*, *Arthrobacter paraffineus*, *Acinetobacter calcoaceticus*, *Alcaligenes* spp., *Acinetobacter cerificans*, etc. (Stepanova *et al.*, 2022). Petroleum hydrocarbon contaminants are less available to these hydrocarbon degraders as they bind to soil particles, which reduces the rate of degradation. When such petroleum contaminated soil is provided with either SACs or microorganisms producing SACs, it shows enhanced degradation of pollutants. Surface active compounds either emulsify or increase surface area of insoluble hydrocarbon pollutants in water to make them available to microorganisms (Ron and Rosenberg, 2002). Rahman *et al.* (2003) evaluated the effect of rhamnolipid SAC on n-alkane utilization by a bacterial consortium. Micelles formed by rhamnolipids solubilize hydrophobic components within them to make them available to microbial cells. These micelles also increase the porosity of cell membrane to facilitate transport of polycyclic aromatic compounds inside. In some cases, biosurfactants have increased membrane hydrophobicity and made direct contact between cells and polycyclic aromatic compounds.

Microbial enhanced oil recovery (MEOR)

MEOR is one of the tertiary oil regaining methods which uses microorganisms as well as various biomolecules produced by them, such as biopolymers, biosurfactants, bioenzymes and biogases, to alter the flow properties of remaining oil in reservoirs using solvents and biogenic acids for improved oil recovery (Bo *et al.*, 2022). Biosurfactants are surface active amphipathic molecules with hydrophobic and hydrophilic fractions both. With this ability, the hydrophilic and aquaphobic parts will solubilize in water and oil phases, respectively. Because of this property, biosurfactant reduces surface tension at the crossing point of oil and water; it also changes the wetting ability and emulsification of crude oil, ultimately eliminating the repulsive forces between oil and water (Rosenberg and Ron, 1999). It has been studied that at low concentrations biosurfactants disperse the crude oil in small particles, while at high concentrations, biosurfactants form micelles with oil and stabilize it. On the other hand, high molecular weight SACs, called as bioemulsifiers, are good at emulsification of oil (Sharma and Pandey, 2020). MEOR is not dependent on usage of harsh chemicals and a high amount of energy, unlike the conventional enhanced oil recovery (EOR) methods. This reduction in chemical usage and energy efficiency lowers environmental contamination and carbon footprint. As MEOR utilizes natural microbial processes, it minimally disturbs ecosystems (Liu and Wei, 2017). By considering all of the above points, it can be concluded that MEOR is the utmost cost-beneficial and environmentally sustainable oil recovery methods. Li and McInerney (2017) have discussed various field trials of oil recovery by using biosurfactant in their book chapter. It summarizes the attempts of Youseff *et al.*, 2007; Joshi *et al.*, 2008; Banat *et al.*, 2014 to inject ex situ synthesized biosurfactants. This book chapter also summarizes injection of microbial biosurfactant producers for greater oil recovery, such as, *Pseudomonas aeruginosa* and *Bacillus subtilis subsp. spizizenii*. Biostimulation of *Pseudomonas*, *Alcaligenes*, and *Rhodococcus* has also been explained.

Bioremediation of heavy metals

Heavy metals are being disposed into the nature because of the anthropogenic events like ore processing, mining, leather tanning, and chemical use. Aquatic ecosystems are getting contaminated with toxic metals like cadmium, nickel, lead, zinc, arsenic, and mercury because of these anthropogenic activities. Industrial operations, use of chemically originated pesticides and fertilizers in agriculture, and non-point source pollution are the reason behind heavy metal pollution (Hama *et al.*, 2023). One of the microbial surface active compounds known as biosurfactant have been effectively applied for the detoxification and/or removal of harmful heavy metals. Surfactants are amphipathic molecules that are often used to solubilize, form a complex, desorb, and mobilize insoluble contaminants into liquid solutions so that bacteria can utilize them. Furthermore, due to their minute size, less toxic impact, and ability to function at varied pH and temperature range, biosurfactants provide clear benefits in redressing heavy metals. A biosurfactant– metal complex is formed as biosurfactants possess a solid affinity intended for heavy metals. Their distinct characteristics, such as their ability to create micelles as well as reduce surface tension, improve the availability and utilization of heavy metals. Therefore, they are essential for cleaning up environmental sites contaminated with heavy metals (Mishra *et al.*, 2021). Mulligan *et al.* (2001) has shown that rhamnolipids produced by *Pseudomonas aeruginosa*, surfactin synthesized by *Bacillus subtilis* and sophorolipid produced by *Torulopsis bombicola* has potential to remove organically bound copper and oxide bound zinc. Gomaa and Meihy, (2019) had found that the biosurfactant formed by *Citrobacter freundii* can remove heavy metals such as aluminum, lead, zinc, cadmium, iron, copper, and manganese. Silva *et al.* (2022) have collectively mentioned about using various biosurfactants to remove heavy metals from environment. Some of the examples are, remediation of chromium using biosurfactant produced by *Bacillus* sp. MTCC 5514, removal of lead, zinc, iron with the help of biosurfactant from *Candida sphaerica*, removal of Cu, Zn, Cr, Pb, Ni and Mn from sludge by using combination of rhamnolipids and saponin.

Removal of chlorinated compounds

Water insoluble chlorinated compounds such as chlorophenols, dichlorobenzenes, and chloroform have significant impact on environment as they are bioaccumulated in aquatic organisms. Many anthropogenic activities such as water treatment practices, chemical manufacturing, and various industrial processes are responsible for release of these chlorinated compounds in aquatic ecosystems (Kuchangi *et al.*, 2023). Application of biosurfactants in removing these chlorinated compounds plays a vital role. These surface active compound enhances the solubilization and dispersion of chlorinated compounds in water. Because of biosurfactants chlorinated compounds are made available to microorganisms, utilizing them and leading to effective remediation of water contaminated with these pollutants (Arman *et al.*, 2021). Biosurfactants such as saponins and rhamnolipids in remediation of polychlorinated biphenyls (Lászlová *et al.*, 2018), biosurfactants from *Rhodococcus* in degradation of chlorinated benzenes (Rapp and Gabriel-Jurgens, 2003), and lipopeptide biosurfactant in the degradation of aromatic amine 4-Chloroaniline (Carolin *et al.*, 2021) demonstrates the effective use of biosurfactants in removal of chlorinated pollutants.

Detergent pollution control

Surfactant is a key component of detergent as it is responsible for active cleaning through penetrating and wetting fabric, loosening soil, and emulsifying soil. Chemical surfactants derived from petrochemicals are being used in detergents mostly. Chemically synthesised surfactants such as ethoxylated fatty acids, alkyl benzenesulfonate, soaps, etc. shown to have critical toxic effects on fresh water creatures. Nearly half of the chemical surfactants synthesized are used in laundry and cleaning sectors. So, biosurfactant with less toxic impact, lower carbon footprint, active at extreme physicochemical conditions and high biodegradability is generating interest in the detergent formulation (Mukharjee and Das, 2010). A recent study on cyclic lipopeptide (CLP) biosurfactants synthesized by *Bacillus subtilis* strain displayed that it was unchanging over a pH variety of 7 – 12 and at 80 ^{O}C for 60 min. It also emulsified efficiently vegetable oils, favouring its inclusion in laundry detergents formulations (Mukharjee, 2007). Less-foaming sophorolipid synthesized by *C. bombicola* seem appropriate for detergent formulation due to its high detergence ability, low toxicity on cells and high degradability and environmental sustainable properties (Hirata *et al.*, 2009). The detergent formulated with rhamnolipid produced by *Pseudomonas aeruginosa* had comparable stain removal ability to commercial powders (Bafghi and Fazaelipoor, 2012). Sajna *et al.*, (2013) also found that the glycolipid biosurfactants isolated from *Pseudozyma* sp. NII08165 was stable at high temperature and alkaline pH. It also showed good stain removal efficiency.

Conclusion

Surface active compounds, particularly bio surfactants and bio emulsifiers, are significantly beneficial for environmental betterment as they improve bioremediation processes. Replacing chemical surfactants with bio surfactants controls water pollution. Practices like microbial enhanced oil recovery support environmental sustainability. Microbial surface active compounds are less toxic, easily biodegradable, highly selective, active at extreme pH, temperature, and saline conditions, and easily synthesized on cheap renewable resources. These characteristics of microbial SACs give them better environmental competence.

Conflict of Interest- None

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