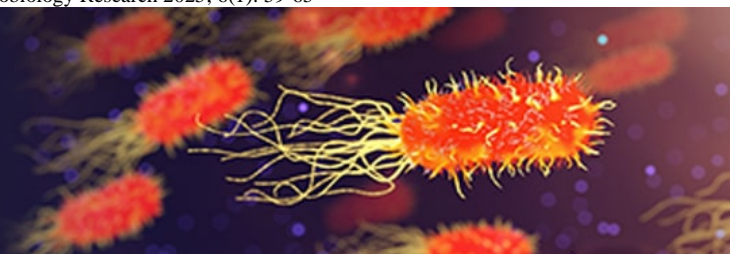


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Biosurfactant-producing bacteria and their applications in various fields of bioremediation

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Abstract

Biosurfactants are surface-active compounds produced by microorganisms, particularly bacteria, with diverse applications in bioremediation. These biomolecules exhibit excellent emulsification, surface tension reduction, and biodegradability, making them eco-friendly alternatives to synthetic surfactants. Various bacterial genera, including *Pseudomonas*, *Bacillus*, and *Rhodococcus*, are well-known biosurfactant producers. Their production is influenced by environmental factors such as nutrient availability, pH, and temperature.

Biosurfactants play a crucial role in bioremediation by enhancing the bioavailability of hydrophobic pollutants, thereby facilitating their degradation. They are widely employed in the remediation of oil spills, heavy metal detoxification, and wastewater treatment. Additionally, biosurfactants contribute to microbial-enhanced oil recovery (MEOR) and the bioremediation of polycyclic aromatic hydrocarbons (PAHs). Their antimicrobial and antifouling properties further extend their applications to environmental and industrial sectors.

The sustainable production of biosurfactants using renewable resources and industrial waste has gained attention, reducing production costs and promoting eco-friendly approaches. Advances in genetic engineering and fermentation technologies are improving biosurfactant yields for large-scale applications. This review highlights the potential of biosurfactant-producing bacteria in bioremediation and discusses recent developments in their production and application strategies.

Keywords: Biosurfactants producing bacteria, bioremediation applications

Introduction

Biosurfactants are amphiphilic compounds synthesized by microorganisms that lower surface and interfacial tension, enhancing the bioavailability of pollutants (Banat *et al.*, 2010) [1]. Unlike synthetic surfactants, they are biodegradable and stable under extreme conditions, making them ideal for bioremediation-the biological degradation or detoxification of environmental contaminants (Mulligan, 2009) [17]. This review focuses on biosurfactant-producing bacteria, their diversity, and their applications across bioremediation fields.

Bioremediation has emerged as a sustainable and eco-friendly approach to mitigate environmental pollution caused by industrialization and anthropogenic activities. Among the various biotechnological tools available for bioremediation, biosurfactants have gained significant attention due to their unique physicochemical properties and broad-spectrum applications. Biosurfactants are amphiphilic compounds produced by various microorganisms, including bacteria, fungi, and yeast, which reduce surface and interfacial tension between two immiscible phases (Banat *et al.*, 2010) [1]. The ability of biosurfactants to enhance the bioavailability of hydrophobic pollutants makes them invaluable in environmental cleanup processes, particularly in the remediation of hydrocarbons, heavy metals, and other toxic contaminants.

Biosurfactants are classified into various groups based on their chemical composition, including glycolipids, lipopeptides, phospholipids, polymeric, and particulate biosurfactants (Mulligan, 2009) [17]. These microbial surfactants exhibit high surface activity, biodegradability, and low toxicity, making them ideal candidates for applications in bioremediation. The advantages of biosurfactants over synthetic surfactants include their effectiveness under extreme environmental conditions such as high salinity, temperature, and pH fluctuations, which further enhance their utility in diverse ecological niches.

The increasing industrial activities and accidental oil spills have led to severe contamination of soil and water resources, posing a significant threat to human health and biodiversity. Traditional remediation methods, such as physical and chemical approaches, often fail to provide a sustainable solution due to high costs, secondary pollution, and limited effectiveness in degrading complex pollutants. In this context, biosurfactant-producing bacteria play a pivotal role in microbial-enhanced oil recovery (MEOR), hydrocarbon degradation, and heavy metal bioremediation by facilitating emulsification, solubilization, and desorption of pollutants from contaminated sites.

Biosurfactant-producing bacteria are predominantly found in hydrocarbon-contaminated environments such as oil spills, petroleum refineries, and industrial wastewater. Common bacterial genera capable of producing biosurfactants include *Pseudomonas*, *Bacillus*, *Rhodococcus*, *Acinetobacter*, and *Serratia*. These bacteria utilize hydrocarbons and other organic substrates as carbon and energy sources, producing biosurfactants as secondary metabolites to enhance substrate solubilization and uptake. The potential of biosurfactant-producing bacteria in environmental remediation has been extensively explored in recent decades, leading to advancements in biotechnological applications.

One of the key applications of biosurfactants in bioremediation is the treatment of petroleum hydrocarbons, which constitute a major class of environmental pollutants. Hydrocarbon-degrading bacteria, often equipped with biosurfactant-producing capabilities, enhance the emulsification of hydrophobic hydrocarbons, thereby increasing their accessibility to microbial enzymatic degradation. The use of biosurfactants in oil spill remediation is particularly relevant for marine environments, where their surface-active properties aid in dispersing oil slicks and facilitating biodegradation by indigenous microbial communities (Banat *et al.*, 2010) ^[1]. Moreover, biosurfactants can be employed in ex-situ and in-situ bioremediation strategies to improve hydrocarbon degradation in contaminated soil and groundwater systems. Apart from hydrocarbon degradation, biosurfactants play a crucial role in heavy metal bioremediation. Heavy metals such as lead, cadmium, mercury, and chromium pose serious environmental and health hazards due to their toxicity, persistence, and bioaccumulation potential. Biosurfactants have demonstrated efficacy in metal sequestration through mechanisms such as chelation, complexation, and bioaccumulation, leading to enhanced metal solubilization and removal from contaminated matrices (Mulligan, 2009) ^[17]. The application of biosurfactant-producing bacteria in heavy metal remediation has been explored in various industrial effluents, mining sites, and wastewater treatment systems, highlighting their potential for sustainable metal detoxification.

Furthermore, biosurfactants exhibit promising applications in bioremediation of emerging pollutants such as pesticides, pharmaceuticals, and microplastics. The hydrophobic nature of many pesticides limits their degradation in soil and water environments. Biosurfactants enhance pesticide bioavailability by increasing their solubility and promoting microbial degradation pathways. Similarly, in the case of pharmaceutical contaminants, biosurfactants facilitate the breakdown of recalcitrant compounds, reducing their persistence in aquatic ecosystems. Recent studies have also

indicated the potential of biosurfactants in the degradation of microplastics, which have become a growing environmental concern due to their widespread distribution and resistance to natural degradation processes.

The industrial production and commercialization of biosurfactants have been steadily increasing due to their multifaceted applications. However, large-scale production of biosurfactants faces several challenges, including high production costs, low yields, and complex downstream processing. Strategies such as metabolic engineering, optimization of fermentation conditions, and utilization of low-cost agro-industrial waste substrates have been explored to enhance biosurfactant yields and reduce production costs. Advances in synthetic biology and genetic engineering have also contributed to improving biosurfactant production by engineering bacterial strains with enhanced biosynthetic pathways.

Biosurfactant-producing bacteria represent a promising tool for sustainable bioremediation of environmental pollutants. Their ability to degrade hydrocarbons, remove heavy metals, and enhance microbial degradation of emerging contaminants underscores their significance in modern environmental biotechnology. Continued research and technological advancements in biosurfactant production, characterization, and application will pave the way for their widespread implementation in environmental cleanup efforts.

Biosurfactant-Producing Bacteria

Classification of Biosurfactants

Biosurfactants are classified by structure:

Glycolipids

Rhamnolipids (*Pseudomonas aeruginosa*)

Glycolipids are one of the most well-studied classes of biosurfactants, known for their excellent surface activity and biodegradability. Among them, rhamnolipids are produced primarily by *Pseudomonas aeruginosa*. Rhamnolipids consist of one or two rhamnose sugar molecules linked to β -hydroxy fatty acid chains. These biosurfactants possess strong emulsifying and antimicrobial properties, making them useful in various industrial applications, including bioremediation, agriculture, and pharmaceuticals. They have been reported to reduce surface tension significantly, enhancing oil recovery and aiding in the bioremediation of hydrocarbons (Santos *et al.*, 2016) ^[25]. Additionally, rhamnolipids play a role in bacterial motility, biofilm formation, and virulence, contributing to the adaptability of *P. aeruginosa* in different environments.

Lipopeptides: Surfactin (*Bacillus subtilis*)

Lipopeptides are cyclic molecules composed of a lipid tail and a short peptide sequence, known for their potent surface activity and antimicrobial effects. One of the most well-characterized lipopeptides is surfactin, produced by *Bacillus subtilis*. Surfactin is a heptapeptide linked to a β -hydroxy fatty acid, forming a cyclic lipopeptide structure. It is highly effective in reducing surface tension and exhibits strong emulsification properties. Surfactin is widely studied for its antimicrobial, antiviral, and anticancer properties. Additionally, it has been applied in environmental cleanup processes, such as the biodegradation of hydrocarbons and heavy metals (Nitschke & Costa, 2007) ^[19]. Its ability to disrupt cell membranes and inhibit biofilm formation makes

it a promising candidate for pharmaceutical and biotechnological applications.

Polymeric Biosurfactants: Emulsan (*Acinetobacter calcoaceticus*)

Polymeric biosurfactants are high-molecular-weight compounds known for their exceptional emulsifying properties. Emulsan, a well-known polymeric biosurfactant, is produced by *Acinetobacter calcoaceticus* and is composed of a polysaccharide backbone with covalently linked fatty acids. Emulsan has been extensively studied for its ability to stabilize oil-in-water emulsions, making it valuable in industries such as petroleum recovery, cosmetics, and food processing. Due to its strong emulsification capacity, it has been used in wastewater treatment and bioremediation efforts to break down hydrophobic contaminants (Ron & Rosenberg, 2001) [23]. Additionally, Emulsan has shown potential applications in drug delivery systems due to its biocompatibility.

Phospholipids and Fatty Acids: Produced by *Acinetobacter* and *Corynebacterium* spp

Phospholipids and fatty acids are another important class of biosurfactants, often found in microbial membranes and involved in surface activity. Certain bacteria, such as *Acinetobacter* and *Corynebacterium* spp., are known to produce phospholipid-based biosurfactants. These molecules consist of hydrophilic phosphate groups attached to hydrophobic fatty acid chains. The biosurfactants derived from these bacteria have been shown to reduce surface tension and exhibit emulsifying properties, making them effective in bioremediation and enhanced oil recovery (Desai & Banat, 1997) [1]. Phospholipids also play a critical role in microbial adaptation to hydrophobic environments and are involved in membrane stability and fluidity.

Key Bacterial Genera Involved in Bioremediation

- *Pseudomonas* species, particularly *Pseudomonas aeruginosa*, produce rhamnolipids, which are potent biosurfactants that enhance hydrocarbon emulsification, making them effective in oil spill remediation and bioremediation of contaminated sites (Pacwa-Płociniczak *et al.*, 2011) [20].
- *Bacillus* species, such as *Bacillus subtilis* and *Bacillus licheniformis*, produce surfactin, a powerful cyclic lipopeptide that aids in the remediation of heavy metals and petroleum hydrocarbons, contributing to wastewater treatment and oil spill cleanup (Singh & Cameotra, 2014) [29].
- *Rhodococcus* species, including *Rhodococcus erythropolis*, synthesize trehalolipids, which facilitate the degradation of persistent polycyclic aromatic hydrocarbons (PAHs) like benzo[a]pyrene and phenanthrene, making them valuable in bioremediation of industrial pollutants (Kuyukina *et al.*, 2015) [11].
- *Acinetobacter* species, such as *Acinetobacter calcoaceticus*, produce emulsan, an extracellular polysaccharide biosurfactant that stabilizes oil-in-water emulsions, making it highly effective in oil spill dispersion and cleanup efforts (Zhang *et al.*, 2012) [33]. Collectively, these biosurfactant-producing microorganisms play a crucial role in environmental bioremediation by enhancing pollutant degradation and promoting sustainable cleanup of contaminated ecosystems.

Production Mechanisms: Biosurfactant synthesis is regulated by specific genes such as *rhlAB* in *Pseudomonas* and *sfp* in *Bacillus*, which are essential for rhamnolipid and Surfactin production, respectively (Chen *et al.*, 2017) [4]. These genes are often controlled by quorum sensing and other regulatory pathways, ensuring biosurfactant production occurs under favourable conditions.

Production is substrate-induced, with hydrocarbons, vegetable oils, and industrial waste serving as key inducers. Factors like pH, temperature, and carbon-nitrogen ratios significantly influence yield, with optimal conditions typically being a slightly alkaline pH and moderate temperatures (Karanth *et al.*, 1999) [9].

Mechanisms of Biosurfactants in Bioremediation

Emulsification: Increases Pollutant Surface Area

Emulsification is a critical mechanism in bioremediation, where biosurfactants enhance the dispersion of hydrophobic pollutants into fine droplets within an aqueous medium, thereby increasing their surface area and bioavailability (Cameotra & Makkar, 2010; Urum & Pekdemir, 2004) [3, 31]. This process reduces interfacial tension, making hydrocarbons more accessible for microbial degradation, which is especially useful in oil spill clean-ups and hydrocarbon bioremediation.

Solubilization: Enhances Solubility of PAHs

Polycyclic aromatic hydrocarbons (PAHs) are persistent pollutants with low aqueous solubility, limiting their natural degradation. Biosurfactants increase PAH solubility by forming micelles, where hydrophobic PAH molecules partition into the surfactant's hydrophobic core, enhancing their dissolution in water (Liu *et al.*, 2018; Cameotra & Makkar, 2010) [13]. This process improves PAH mobility and bioavailability for microbial degradation, making it a promising approach for remediating PAH-contaminated environments.

Desorption: Releases Contaminants from Soil

Contaminants such as petroleum hydrocarbons and heavy metals often adhere strongly to soil particles, reducing their bioavailability. Biosurfactants facilitate desorption by decreasing the adhesion forces and interfacial tension between contaminants and soil matrices (Urum & Pekdemir, 2004; Liu *et al.*, 2018) [13, 31]. This process enhances pollutant mobility, improving the efficiency of soil washing and bioremediation techniques.

Metal Chelation: Binds Metals like Pb and Cd

Heavy metals like lead (Pb) and cadmium (Cd) pose significant environmental and health risks due to their non-biodegradable nature. Biosurfactants containing anionic functional groups (e.g., carboxyl, hydroxyl) can chelate metal ions, forming stable complexes that reduce their toxicity and facilitate their removal (Mulligan *et al.*, 2001; Urum & Pekdemir, 2004) [18, 31]. This mechanism is crucial in enhancing phytoremediation and metal recovery from contaminated sites.

Applications in Bioremediation

Hydrocarbon Degradation by Biosurfactants

Hydrocarbon pollution, primarily from crude oil spills, poses a significant environmental threat. The removal of these pollutants is often challenging due to the hydrophobic

nature of hydrocarbons, which limits their bioavailability to microorganisms. Biosurfactants, amphiphilic compounds produced by microbes, enhance the solubility and emulsification of hydrocarbons, facilitating microbial degradation. Among the various biosurfactants studied, rhamnolipids from *Pseudomonas aeruginosa* and surfactin from *Bacillus subtilis* have shown promising results in hydrocarbon degradation.

Rhamnolipids and Hydrocarbon Biodegradation

Rhamnolipids, glycolipid biosurfactants primarily produced by *Pseudomonas aeruginosa*, play a crucial role in crude oil degradation. These biosurfactants reduce surface and interfacial tension, promoting the emulsification of hydrocarbons and enhancing microbial access to oil contaminants. According to Rahman *et al.* (2003) [22], rhamnolipids facilitated the degradation of crude oil in contaminated soil, achieving 70% removal efficiency. This effect is attributed to the increased bioavailability of hydrocarbons to oil-degrading microbial communities. Additionally, rhamnolipids have been shown to enhance microbial adhesion to hydrocarbon surfaces, further promoting degradation. Their ability to withstand extreme environmental conditions, such as high salinity and temperature, makes them ideal for in situ bioremediation applications.

Surfactin and Alkane Degradation

Surfactin, a cyclic lipopeptide biosurfactant produced by *Bacillus subtilis*, has been extensively studied for its role in hydrocarbon degradation. Thavasi *et al.* (2011) [30] reported that surfactin enhanced alkane degradation by 60%, demonstrating its potential in the bioremediation of petroleum hydrocarbons. Surfactin's strong emulsification properties allow the dispersion of hydrophobic hydrocarbons, facilitating microbial uptake and metabolism. Furthermore, surfactin-producing bacteria exhibit synergistic interactions with hydrocarbon-degrading microbes, leading to more efficient pollutant breakdown. The addition of surfactin to contaminated environments has been proposed as a strategy to improve the biodegradation efficiency of naturally occurring microbial communities.

Role of Biosurfactants in Heavy Metal Remediation

Biosurfactants facilitate heavy metal removal by mechanisms such as chelation, complexation, ion exchange, and enhanced solubilization. These compounds can alter metal bioavailability, making them easier to extract from contaminated soils, sediments, and wastewater.

Surfactin and Its Role in Heavy Metal Removal

Surfactin is a cyclic lipopeptide biosurfactant produced by *Bacillus subtilis*. It has been reported to effectively bind and remove cadmium (Cd) and lead (Pb) from contaminated soil.

- Mulligan & Gibbs (2004) demonstrated that surfactin achieved up to 65% removal of Cd and Pb from soil.
- The removal mechanism involves metal chelation and adsorption onto micelles, thereby reducing metal toxicity and mobility.
- Surfactin also enhances metal bioavailability for further degradation or extraction.

Rhamnolipids and Their Effect on Zinc Mobilization

Rhamnolipids are glycolipid biosurfactants mainly produced

by *Pseudomonas aeruginosa*. They are particularly effective in mobilizing metals in contaminated sediments.

- Herman *et al.* (1995) [8] reported that rhamnolipids significantly enhanced the mobilization of zinc (Zn) in sediments.
- Rhamnolipids work by reducing surface tension and forming stable metal complexes, allowing Zn to be desorbed from soil particles and solubilized into the aqueous phase.
- These biosurfactants also increase metal uptake by microorganisms and plants, promoting bioremediation through bioaccumulation and phytoremediation.

Advantages of Biosurfactants in Metal Remediation

- Eco-friendly and Biodegradable - Unlike synthetic surfactants, biosurfactants do not persist in the environment and degrade naturally.
- Highly Effective at Low Concentrations - Even at low concentrations, biosurfactants can enhance metal solubilization and removal.
- Compatible with Other Bioremediation Techniques - They can be combined with microbial remediation and phytoremediation for enhanced efficiency.
- Cost-Effective - Can be produced using industrial waste as substrates, reducing production costs.

Soil Remediation: Microbial Approaches for Contaminant Degradation:

Soil pollution caused by organic contaminants such as polycyclic aromatic hydrocarbons (PAHs) and pesticides poses significant environmental and agricultural challenges. Bioremediation, an eco-friendly and sustainable approach, utilizes microorganisms and their metabolic products to degrade or remove these pollutants from soil. Among the various microbial strategies, *Rhodococcus* spp. and biosurfactants have shown remarkable efficiency in soil remediation.

Role of *Rhodococcus* spp. in PAH Degradation

Polycyclic aromatic hydrocarbons (PAHs) are persistent organic pollutants primarily released from fossil fuel combustion, industrial activities, and oil spills. These compounds are highly hydrophobic and pose a risk to soil health and groundwater contamination. *Rhodococcus* spp., a genus of actinobacteria, has been widely studied for its ability to degrade PAHs due to its versatile metabolic pathways and enzymatic systems.

According to Martinez *et al.* (2015) [15], *Rhodococcus* spp. exhibit an 85% degradation efficiency in breaking down PAHs, making them highly effective bioremediation agents. These bacteria utilize oxygenase enzymes such as monooxygenases and dioxygenases, which catalyze the oxidation of PAHs into less toxic intermediates. The high adaptability of *Rhodococcus* spp. to harsh environmental conditions, including extreme pH, temperature, and salinity, further enhances their bioremediation potential.

Biosurfactants in Pesticide Removal

Pesticide residues in agricultural soils can lead to soil toxicity, reduced microbial diversity, and contamination of water resources. Conventional remediation techniques, such as chemical treatments, may not be efficient and can introduce secondary pollutants. Biosurfactants, microbial surface-active compounds, offer a sustainable alternative by

enhancing the solubilization and biodegradation of pesticides.

Fenibo *et al.* (2019) ^[6] reported that biosurfactants significantly improve pesticide removal from soil by increasing the bioavailability of hydrophobic contaminants. Biosurfactants reduce surface tension and emulsify hydrophobic pollutants, facilitating their microbial uptake and degradation. Some biosurfactant-producing microorganisms, such as *Pseudomonas* and *Bacillus* species, can simultaneously degrade pesticides and produce biosurfactants, enhancing the efficiency of bioremediation.

Wastewater Treatment Using Biosurfactants

Wastewater treatment is a crucial process for reducing pollutants and improving water quality before discharge or reuse. Industrial effluents often contain high levels of organic pollutants, oils, and heavy metals, which can be challenging to remove using conventional treatment methods. Biosurfactants, due to their surface-active properties, have emerged as eco-friendly and efficient alternatives for enhancing wastewater treatment.

Role of Biosurfactants in Wastewater Treatment

Reduction of Chemical Oxygen Demand (COD)

Chemical Oxygen Demand (COD) is a key indicator of water pollution, representing the amount of oxygen required to oxidize organic and inorganic matter in wastewater. High COD levels indicate severe contamination, which can harm aquatic life and the environment. Emulsan, a microbial biosurfactant, has been reported to reduce COD by approximately 60% in industrial effluents (Kosaric, 2001) ^[10]. This reduction is attributed to Emulsan's emulsifying properties, which enhance the breakdown and removal of hydrophobic pollutants from wastewater.

Oil Removal from Wastewater

Industrial wastewater, particularly from petroleum, food processing, and chemical industries, often contains oil and grease, making it difficult to treat using conventional methods. Biosurfactants such as surfactin, produced by *Bacillus* species, have been shown to effectively aid oil removal. Surfactin disrupts oil-water interfaces, facilitating the emulsification and subsequent degradation of hydrocarbons. Studies have demonstrated that surfactin enhances oil dispersion and biodegradation in wastewater treatment systems (Wei *et al.*, 2016) ^[32].

Heavy Metal Removal

Biosurfactants also play a significant role in the removal of heavy metals from wastewater. They form complexes with metal ions, enhancing their solubility and bioavailability for microbial uptake and degradation. Rhamnolipids, produced by *Pseudomonas aeruginosa*, have been widely studied for their potential in heavy metal remediation, effectively mobilizing metals such as cadmium, lead, and zinc.

Biodegradability and Environmental Benefits

Unlike synthetic surfactants, biosurfactants are biodegradable, non-toxic, and environmentally friendly. Their use in wastewater treatment reduces dependency on chemical surfactants, which can contribute to secondary pollution. Additionally, biosurfactants are effective under a wide range of pH, temperature, and salinity conditions, making them suitable for various industrial effluents.

Marine Bioremediation and the Role of *Alcanivorax* Biosurfactants in Oil Degradation

Marine bioremediation is an eco-friendly approach that utilizes microorganisms to degrade and remove pollutants from marine environments. One of the most critical applications of marine bioremediation is in the cleanup of oil spills, which pose significant environmental threats to aquatic ecosystems. Among the various oil-degrading microorganisms, *Alcanivorax* species play a crucial role due to their ability to produce biosurfactants that aid in the biodegradation of hydrocarbons.

Role of *Alcanivorax* in Oil Degradation

Alcanivorax is a genus of marine bacteria known as hydrocarbonoclastic bacteria (HCB), which thrive in oil-contaminated environments by utilizing hydrocarbons as their primary source of carbon and energy. When oil spills occur, these bacteria proliferate rapidly and contribute to the natural degradation of petroleum hydrocarbons in seawater.

Alcanivorax Biosurfactants and Their Function

Biosurfactants are surface-active molecules produced by microorganisms that help in emulsifying hydrophobic compounds like crude oil. The biosurfactants produced by *Alcanivorax* serve multiple functions in marine bioremediation:

Oil Dispersion: The biosurfactants reduce the surface tension between oil and water, leading to the breakdown of large oil slicks into smaller droplets. This enhances the bioavailability of hydrocarbons for microbial degradation.

Emulsification of Hydrocarbons: By increasing the solubility of hydrophobic petroleum compounds, biosurfactants allow microbial enzymes to access and degrade hydrocarbons more efficiently.

Enhanced Biodegradation Rates: The presence of biosurfactants accelerates the microbial breakdown of oil pollutants, reducing the persistence of hydrocarbons in the marine ecosystem.

According to Hassanshahian *et al.* (2014) ^[7], *Alcanivorax* biosurfactants play a significant role in dispersing oil slicks, thereby enhancing the degradation of petroleum hydrocarbons. This process significantly contributes to the natural self-cleaning mechanisms of the ocean following oil spills.

Environmental Implications

The use of *Alcanivorax* and its biosurfactants in marine bioremediation presents an effective and sustainable solution for mitigating oil pollution. Compared to chemical dispersants, biosurfactants are biodegradable, non-toxic, and environmentally friendly. Their application in oil spill cleanup strategies can minimize long-term ecological damage and support the restoration of marine habitats.

Advantages and Challenges

Advantages: Biodegradable and Low Toxicity

Biosurfactants are amphiphilic compounds produced by microorganisms, including bacteria and fungi that possess excellent surface-active properties. Unlike synthetic surfactants, biosurfactants are biodegradable, meaning they can be broken down naturally by microbial activity in the

environment, reducing long-term ecological impact. Their low toxicity makes them suitable for various applications, including bioremediation, pharmaceuticals, cosmetics, and food industries. According to Satpute *et al.* (2010) ^[26], biosurfactants demonstrate minimal adverse effects on human health and the ecosystem, making them a sustainable alternative to chemically synthesized surfactants.

Effective at Low Critical Micelle Concentration (CMC)

The critical micelle concentration (CMC) is the minimum concentration at which surfactant molecules aggregate to form micelles, which is crucial for reducing surface and interfacial tension. Biosurfactants, such as rhamnolipids and lipopeptides, are highly effective even at low CMC values compared to synthetic surfactants. Lin *et al.* (2013) ^[12] reported that biosurfactants require significantly lower concentrations to achieve similar or superior surface tension reduction, making them more efficient in industrial applications like enhanced oil recovery, detergents, and wastewater treatment.

Stable Under Extreme Conditions

One of the remarkable features of biosurfactants is their stability under extreme environmental conditions, including high salinity, extreme pH, and elevated temperatures. This characteristic makes them highly suitable for industrial applications where harsh conditions often degrade synthetic surfactants. For instance, Perfumo *et al.* (2010) ^[21] demonstrated that certain biosurfactants, such as glycolipids and lipopeptides, remain functionally active even under extreme temperatures (up to 100°C), acidic or alkaline conditions, and in the presence of high salt concentrations. This stability enhances their effectiveness in bioremediation, oil spill clean-up, and pharmaceutical formulations.

Challenges in Biosurfactant Production and Application

Despite the promising potential of biosurfactant-producing bacteria in bioremediation, several challenges hinder their large-scale production and commercial application. These challenges primarily include high production costs, scalability issues, and regulatory barriers.

High Production Costs

One of the major limitations in the commercialization of biosurfactants is their high production cost. Biosurfactants are primarily produced through microbial fermentation, which requires expensive substrates, specialized equipment, and controlled environmental conditions (Makkar & Cameotra, 2002) ^[14]. Traditional carbon sources such as glucose and glycerol contribute significantly to production costs, making biosurfactants less economically viable compared to synthetic surfactants. Additionally, downstream processing, including extraction and purification, further escalates costs due to the need for solvents and complex separation techniques. To address this issue, researchers are exploring cost-effective production strategies, such as the use of agro-industrial waste (e.g., molasses, whey, and vegetable oils) as alternative substrates to reduce raw material expenses (Banat *et al.*, 2014) ^[2].

Scalability Issues

Another significant challenge is the difficulty in scaling up biosurfactant production from laboratory to industrial levels. While small-scale production in controlled environments

yields promising results, large-scale bioreactors often present issues such as contamination risks, variability in microbial performance, and difficulties in maintaining optimal growth conditions (Rufino *et al.*, 2014) ^[24]. Moreover, biosurfactant production is influenced by factors such as pH, temperature, aeration, and nutrient availability, making process optimization challenging. Industrial-scale production also requires efficient recovery and purification methods, which remain complex and cost-intensive. Innovative bioprocess engineering approaches, including metabolic engineering, bioreactor optimization, and continuous fermentation systems, are being explored to overcome these scalability challenges (Mulligan, 2009) ^[17].

Regulatory Barriers

The commercialization of biosurfactants is further complicated by regulatory challenges. Unlike synthetic surfactants, which have well-established industrial standards, biosurfactants face stringent regulatory approvals before they can be widely used in environmental and industrial applications (Shete *et al.*, 2006) ^[28]. Regulatory bodies require extensive toxicological and environmental impact assessments to ensure biosurfactants are safe for human health and ecosystems. This process can be time-consuming and costly, discouraging companies from investing in large-scale biosurfactant production. Additionally, the lack of standardized protocols for biosurfactant classification and approval further delays commercialization. To accelerate regulatory approvals, collaborative efforts between academia, industry, and policymakers are needed to establish clear guidelines and safety regulations.

Future Prospects

The application of biosurfactant-producing bacteria in bioremediation holds immense potential for sustainable environmental management. Advances in genetic engineering and metabolic pathway optimization can enhance biosurfactant yield and functionality, making them more efficient in oil spill cleanup, heavy metal detoxification, and the degradation of hydrophobic pollutants (Mukherjee *et al.*, 2006) ^[16]. Cost-effective large-scale production using agro-industrial waste as substrates can improve their commercial viability (Banat *et al.*, 2014) ^[2]. The integration of biosurfactants with nanotechnology and bioaugmentation techniques is expected to further enhance their remediation efficiency (Mulligan, 2009) ^[17]. Regulatory approvals and industry collaborations will drive their widespread adoption in wastewater treatment, soil remediation, and green chemistry applications (Satpute *et al.*, 2010) ^[27].

Conclusion

Biosurfactant-producing bacteria, such as *Pseudomonas*, *Bacillus*, and *Rhodococcus*, represent a transformative force in bioremediation, offering sustainable solutions for environmental pollution. Their ability to produce eco-friendly, biodegradable surfactants like rhamnolipids, surfactin, and emulsan enables the efficient degradation of hydrocarbons, removal of heavy metals, and remediation of contaminated soils and waters. These microorganisms enhance pollutant bioavailability through emulsification, solubilization, and desorption, outperforming synthetic alternatives under diverse conditions. Applications span oil

spill cleanup, wastewater treatment, and agricultural soil restoration, demonstrating versatility and efficacy. Despite their promise, challenges such as high production costs and scalability limitations hinder widespread adoption. Advances in genetic engineering and fermentation optimization offer pathways to overcome these barriers, paving the way for large-scale implementation. As environmental concerns escalate, biosurfactants from bacteria stand as a cornerstone of green technology, poised to redefine bioremediation strategies and promote a cleaner, healthier planet.

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