

Biosurfactants in the remediation of petroleum-contaminated soils: Mechanisms and applications

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ABSTRACT

Petroleum contamination of soils persists despite decades of remediation research. Growing evidence positions biosurfactants as practical tools that align clean-up with soil health. This review synthesizes recent laboratory, pilot, and field studies on rhamnolipids, sophorolipids, trehalose lipids, and lipopeptides. Core actions include reduction of surface and interfacial tension, micelle formation, emulsification of hydrocarbons, and desorption from soil particles. These processes increase bioavailability for indigenous degraders and accelerate loss of persistent fractions. Reported co-benefits include improved infiltration in hydrophobic matrices, better chemical balance in saline soils, and shifts in microbial communities that support sustained recovery. Applications span soil washing and in situ delivery, with consistent gains when biosurfactants are integrated with bioaugmentation, phytoremediation, electrokinetics, or engineered biochar. Performance is dose dependent and bounded by a safety window. Excessive loading can stress plants and impair soil biota, which requires stepwise pilots, dose ceilings, and monitoring of biological indicators. Barriers remain in production cost, formulation stability, and supply at scale. Advances in strain engineering, circular feedstocks, and gentler recovery methods are reducing these constraints. Priority needs include standardized protocols, multi-site demonstrations, and evaluation frameworks that track both contaminant removal and soil function. When molecule choice and delivery are matched to site chemistry, biosurfactants can progress from promising additives to cornerstone technologies for sustainable remediation of petroleum-impacted soils.

Keywords: biosurfactants, petroleum-contaminated soils, bioremediation, soil washing, bioavailability.

INTRODUCTION

Petroleum-contaminated soils continue to pose a significant threat to agroecosystems and public health. Conventional remediation methods – such as thermal treatment, solvent extraction, chemical oxidation, and solidification – can reduce hydrocarbon concentrations; however, these approaches are often energy-intensive, costly, and prone to transferring pollutants from one phase to another. As a result of these limitations, attention has increasingly shifted toward biologically based remediation strategies that align with sustainability objectives (Eras-Muñoz et al., 2022).

Biosurfactants are surface-active compounds produced by microorganisms. Owing to their amphiphilic nature, these molecules reduce surface and interfacial tension, disperse hydrophobic compounds, and enhance mass transfer. They are biodegradable, exhibit relatively low toxicity, and retain their activity across broad ranges of salinity, temperature, and pH. Collectively, these characteristics make biosurfactants strong candidates for the remediation of petroleum-impacted soils and the restoration of soil function following treatment (Sharma et al., 2022). Given these advantages, current research focuses on well-studied biosurfactant families – including glycolipids

(such as rhamnolipids, sophorolipids, and trehalose lipids) and lipopeptides (such as surfactin) – as the field transitions from proof-of-concept to practical deployment (Nagtode et al., 2023).

In soil environments, biosurfactants operate via multiple complementary mechanisms. They desorb hydrocarbons from mineral and organic surfaces, emulsify or solubilize non-aqueous phase liquids, and increase the bioavailability of contaminants to indigenous microbial degraders. These effects collectively accelerate both *ex situ* soil washing and *in situ* bioremediation processes. Optimized rhamnolipid washing has achieved approximately 60% reduction in total petroleum hydrocarbons in field soils, with even higher removal observed for selected hydrocarbon fractions; this outcome underscores the importance of remediation designs that account for both contaminant composition and total load (Olasanmi and Thring, 2020). Similarly, sophorolipids have demonstrated comparable improvements in hydrocarbon biodegradation and have been employed as functional additives in electrokinetic remediation systems and biochar-based soil amendments, thereby improving contact among carbonaceous sorbents, microbial communities, and weathered hydrocarbons (Chen et al., 2024).

Early comparative studies have also shown that, when dose and contact time are properly optimized, biosurfactants can rival or outperform synthetic surfactants in washing crude-oil-contaminated soils (Nagtode et al., 2023). Beyond contaminant removal, surfactant-induced changes at the soil–water–air interface can alter soil physical behavior. By modifying wettability and interfacial tension, biosurfactants may reduce water repellency and improve infiltration in hydrophobic soils, potentially aiding recovery of soil hydraulic function after spills or fires. These effects depend on factors such as soil texture, organic matter content, and the specific surfactant chemistry, so they must be evaluated on a case-by-case basis (Tiwari and Tripathy, 2023). Biosurfactant application also influences microbial community composition and enzyme activity in the soil. However, these benefits are strongly dose-dependent: excessive concentrations of biosurfactant can inhibit plant growth or microbial processes, underscoring the need for carefully formulated, site-specific treatments (Markam et al., 2024).

Recent research highlights practical benefits when biosurfactants are integrated with other

remediation strategies, such as bioaugmentation consortia, composting, phytoremediation, or conductive soil amendments. In laboratory soil experiments, combinations of rhamnolipids and lipopeptides with a tailored bacterial inoculum have increased removal of total petroleum hydrocarbons and polycyclic aromatic hydrocarbons by several tens of percentage points compared with inoculation alone, while also reducing ecotoxicity indicators. These outcomes support the core goals of remediation: reducing risk and restoring soil function (Filho et al., 2023). Nonetheless, wider application of biosurfactant-based methods still faces hurdles. Production costs, downstream purification, and scaling up supply remain significant bottlenecks, although advances in alternative feedstocks and intensified production processes are beginning to alleviate these challenges (Solomon and Vishnu, 2025).

Recent reviews predominantly address microbial production/bioprocessing and market aspects or provide broad environmental overviews (Santos et al., 2024; Thakur et al., 2024). Other surveys cover water and generic soil systems without translating mechanisms into soil-specific design rules for petroleum matrices (Eras-Muñoz et al., 2022; Nagtode et al., 2023). Petroleum-soil reviews do exist, but they rarely connect structure/solution behavior to decision-relevant soil metrics or define ecological safety windows (Liu et al., 2021; Parus et al., 2023; Zhao et al., 2024).

In this review, we link structure and solution behavior (head-tail chemistry, CMC, surface and interfacial-tension control, wetting, micellar/emulsion regimes) to quantifiable outcomes across heterogeneous soils, propose a decision matrix for molecule/dose selection, define CMC-normalized dosing and delivery routes, and integrate biosurfactants with complementary remediation strategies within defined ecological safety windows (GI, DHA). Together, these elements provide soil-specific design guidance and clear integration pathways for petroleum-impacted soils.

BIOSURFACTANTS: TYPES AND PROPERTIES

Definition and distinction from chemical surfactants

Biosurfactants are surface-active molecules produced by microorganisms. They are

amphiphiles that lower surface and interfacial tension, form organized assemblies in water, and disperse hydrophobic compounds. In contrast, many synthetic surfactants are petrochemical-derived and may show higher toxicity and slower biodegradation (Simões et al., 2024). Recent classifications group biosurfactants by chemistry into glycolipids and lipopeptides and by producer into bacteria, yeasts, and filamentous fungi. These schemes help link structure to function in soil systems (Sah et al., 2022). The literature also reports wide operational stability across salinity, temperature, and pH, which is valuable in field soils. Sodium dodecyl sulfate is a common synthetic benchmark with a high critical micelle concentration and a moderate surface tension reduction compared with several biosurfactants (Dini et al., 2024).

Main classes used in petroleum-impacted soils

Rhamnolipids. Glycolipids produced mainly by *Pseudomonas* species. They show strong interfacial activity and are widely studied for soil washing and in situ use (Kabeil et al., 2025; Mahamad et al., 2025; Sikandar et al., 2024).

Sophorolipids. Glycolipids produced by *Starmerella bombicola* and related yeasts. They are available at scale, show low toxicity in several models, and are increasingly used in environmental and agricultural formulations (Delbeke et al., 2016; Roelants et al., 2024). **Trehalose lipids.** Glycolipids from *Rhodococcus* and related genera. They reduce surface tension efficiently and support hydrocarbon dispersion and biodegradation (Andreolli et al., 2023; Zhu et al., 2026).

Lipopeptides including surfactin and serrawettin. Cyclic peptide–lipid amphiphiles from *Bacillus* and *Serratia*. Surfactin is among the most potent biosurfactants for lowering surface tension. Serrawettin variants show strong wetting and emulsification and are relevant to hydrocarbon mobilization (Chen et al., 2024; Théâtre et al., 2021).

Key surface properties

Surface and interfacial tension. Rhamnolipids commonly lower water surface tension from about seventy-two millinewtons per meter to near thirty. Interfacial tension between water and hydrocarbons can approach one millinewton per meter under optimized conditions. Sophorolipids lower water surface tension into the thirties at low doses. Surfactin reaches values near twenty-five

to twenty-seven millinewtons per meter. Trehalose lipids often reach the mid-thirties. These ranges are well documented across recent studies and reviews (Dini et al., 2024).

Critical micelle concentration. Rhamnolipid CMC values typically span ten to two hundred thirty milligrams per liter with formulation and ionic strength as key drivers. Increased salinity often lowers CMC for rhamnolipids, which favors micellization in brackish soils. Reported CMC values for sophorolipids vary by chain length and lactone versus acid forms and are generally higher than for rhamnolipids. Trehalose lipids show CMC values reported from twenty-five to one hundred forty milligrams per liter, reflecting producer strain and purity (Zhang et al., 2022).

Stability in soil-like conditions. Multiple studies describe activity across broad pH, salinity, and temperature windows. This supports use in variable field sites and during seasons with fluctuating soil moisture (Ali et al., 2021).

These performance parameters are often visualized through the relationship between surfactant concentration and surface tension. Figure 1 illustrates how biosurfactant monomers reduce surface tension until the critical micelle concentration is reached, beyond which micelles form and stabilize the system. Such behavior explains why rhamnolipids and surfactin are particularly efficient in mobilizing hydrocarbons from soil matrices (Hernández et al., 2023).

Comparative analysis also shows that while rhamnolipids can reach surface tension values near thirty millinewtons per meter, their CMC spans a broad range and may be higher than for trehalose lipids under certain conditions. Sophorolipids, although less potent in reducing surface tension, are attractive because they are scalable and stable. Surfactin provides the lowest values, but its production cost is limiting. Table 1 summarizes representative results for surface tension reduction and CMC values, highlighting trade-offs between efficiency and feasibility across biosurfactant classes.

Environmental advantages

Biosurfactants are readily biodegradable and often show lower aquatic and terrestrial toxicity than many synthetic analogues. They are compatible with living systems, including plant–microbe consortia used in bioremediation. Their production can use renewable feedstocks from

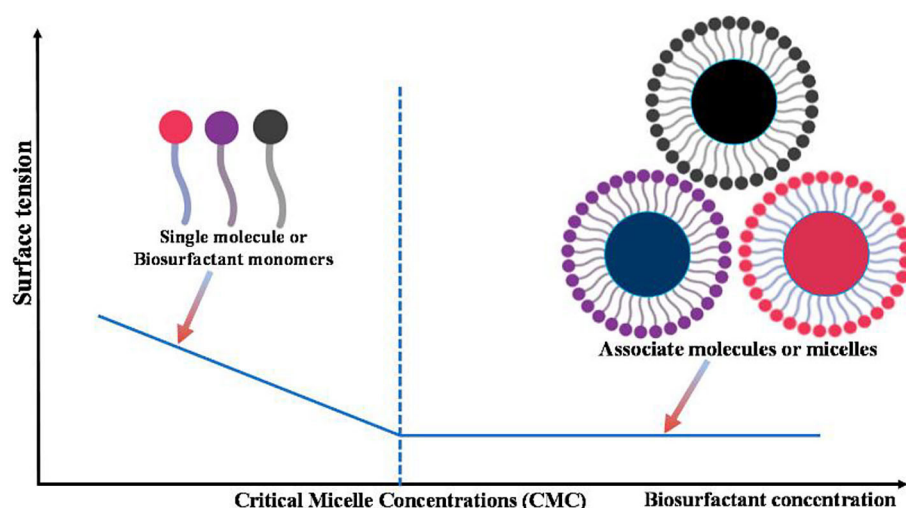


Figure 1. Micelle formation and the concept of critical micelle concentration for a microbial surfactant

Table 1. Key results from recent studies on surface activity

Biosurfactant	Representative surface activity result	Ref.
Surfactin	Water surface tension reduced to about 25–27 mN m ⁻¹ .	(Phulpoto et al., 2020)
Rhamnolipids	Water surface tension reduced from 72 to about 30 mN m ⁻¹ . Water–kerosene interfacial tension down to about 1 mN m ⁻¹ under optimization.	(Kabeil et al., 2025; Zhou et al., 2019)
Rhamnolipids	CMC reported from 10 to 230 mg L ⁻¹ depending on strain, pH, and ionic strength.	(Chong and Li, 2017)
Rhamnolipids	CMC decreases as NaCl increases to a reported minimum near 110 mg L ⁻¹ .	(Zhang et al., 2022)
Sophorolipids	Water surface tension commonly lowered to the 30–40 mN m ⁻¹ range at low dose.	(Delbeke et al., 2016)
Trehalose lipids	Water surface tension near 34 mN m ⁻¹ with CMC around 0.14 mg mL ⁻¹ .	(Janek et al., 2018)
Trehalose lipids	CMC reported near 0.025 g L ⁻¹ in purified preparations.	(Salek et al., 2022)
Serrawettin	Reported strong wetting and emulsification with confirmed biosurfactant identity and production advances.	(Chen et al., 2024)

agro-industrial waste, which improves life-cycle metrics. Sophorolipids in particular have documented safety margins in topical and environmental uses. These attributes align with soil health goals during and after petroleum clean-up (Nag-tode et al., 2023; Patel and Kharawala, 2022).

PETROLEUM CONTAMINATION IN SOILS: CHALLENGES AND MECHANISMS

Petroleum hydrocarbons are persistent in soils because they partition into pore spaces, adsorb onto minerals and organic matter, and display low aqueous solubility. These traits limit mass transfer to microbes and slow natural attenuation. Weathering further increases heterogeneity as residues age and bind strongly to soil matrices. Site conditions such as texture, water status, porosity, and organic carbon control these processes and often dictate remediation outcomes (Mekonnen et al., 2024).

Polycyclic aromatic hydrocarbons represent a major challenge due to low solubility, strong sorption, and toxicity. High-molecular-weight congeners are especially recalcitrant and can persist for years with bioaccumulation risk across food webs. Heavy petroleum fractions such as resins and asphaltenes also weather slowly and resist biodegradation, which sustains long-term soil risk (Patel et al., 2020).

Conventional technologies can reduce pollutant loads yet often at high energy demand or with secondary impacts. Thermal treatments and incineration require substantial heat input. Chemical oxidation can generate undesirable by-products. Even soil washing with synthetic surfactants may shift contaminants to another phase rather than removing risk at the source. These trade-offs explain the growing interest in greener alternatives that increase bioavailability without adding new hazards (Zhang et al., 2024). Figure 2 illustrates the dominant pathways that govern

petroleum behavior in soil, including volatilization of light fractions, dissolution of soluble components, sorption and entrapment within organic matter and micropores, and biological transformation by indigenous microbiota. The schematic clarifies why petroleum remains difficult to mobilize and degrade in the absence of measures that enhance mass transfer and access to bound residues (Wang et al., 2021).

MECHANISMS OF BIOSURFACTANT ACTION IN SOILS

Biosurfactants enhance soil clean-up through physicochemical and biological routes that act together. They lower surface and interfacial tension, form micelles and emulsions, promote desorption from soil particles, and increase mass transfer to degraders. At the same time they modulate cell–surface properties and biofilms, which improves access to hydrocarbons. These coupled effects increase bioavailability and move petroleum residues from poorly accessible domains into degradable pools (Silva et al., 2024). Micellar solubilization and emulsification raise the apparent aqueous solubility of polycyclic aromatic hydrocarbons and other weathered fractions. As micelles form, slowly desorbing pools move into the aqueous phase, which often governs long-term risk in aged soils. Rhamnolipid doses adjusted with ionic strength have repeatedly increased solubilization and subsequent biodegradation in field-impacted soils (Arkhipov et al., 2024).

Lower interfacial tension and shifts in contact angle change soil wettability. Hydrocarbons detach from mineral and organic surfaces, which enables soil washing and in situ flushing. Coupling with electrokinetic delivery moves reagents and mobilized contaminants through low-permeability zones and improves contact at depth (Ogunmokun and Wallach, 2023; Omo-Okoro et al., 2025). At the interface between microbes and hydrocarbons, biosurfactants can modify cell surface hydrophobicity and reorganize biofilms. These changes open transport pathways, improve access to hydrophobic substrates, and increase degradation rates when doses remain within toxicity limits (Vandana and Das, 2023). In soils co-contaminated with metals, anionic biosurfactants can chelate or form complexes with metal cations. Complexation may mobilize metals during washing or reduce their bioavailability when bound to sorbents. Site-specific design and careful sequencing are required when petroleum and metals coexist (Lee et al., 2025).

Hybrid treatment schemes that combine interfacial control with targeted chemical transformation offer further gains: for example, rhamnolipid-mediated elution followed by zero-valent iron-activated persulfate oxidation has removed persistent PAHs more effectively than either mobilization or oxidation alone, illustrating the value of staged interventions that first increase availability and then transform recalcitrant compounds (Guo et al., 2025).

Figure 3 schematically summarizes the main pathways by which biosurfactants act in soils, emphasizing desorption from soil particles, micellar

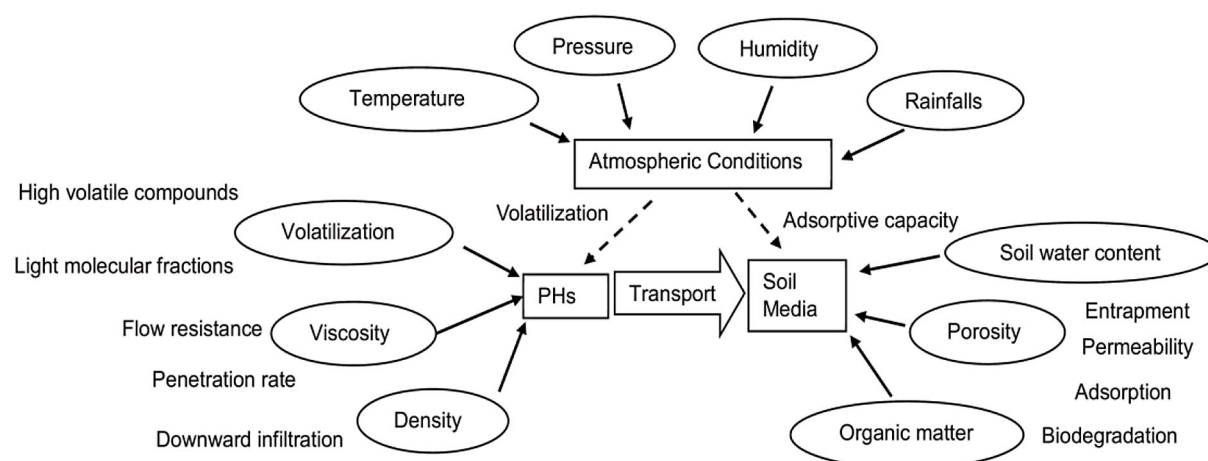


Figure 2. Fate and weathering of petroleum hydrocarbons in soil, showing volatilization, dissolution, sorption–desorption, diffusion into micropores, and biodegradation by native communities

solubilization and emulsification, and enhanced microbial uptake – mechanisms that together increase the bioavailability of petroleum residues for biodegradation (Ambaye et al., 2021).

EFFECTS OF BIOSURFACTANTS ON SOIL PROPERTIES

Biosurfactants can shift key soil properties in ways that support remediation and recovery. By lowering surface and interfacial tension they improve wetting of hydrophobic pores and promote infiltration, which helps re-establish hydraulic continuity after petroleum spills or fire-induced water repellency (Ogunmokun and Wallach, 2023).

In saline and sodic settings, rhamnolipid amendments have improved the soil microenvironment. A recent study reported increases in soil organic matter by five to thirty-one percent and reductions in electrical conductivity by about zero point three to forty-two percent, with concurrent changes in the rhizosphere community that favor stress tolerance (Liu et al., 2023). Field work in cotton systems showed rhamnolipid drip irrigation lowered rhizosphere salinity by roughly ten, five, and three percent in slightly, moderately, and highly salinized soils respectively, with improved plant performance (Chen et al., 2024). These outcomes indicate that biosurfactants can help restore chemical balance and root-zone function when salts co-occur with petroleum residues.

Table 2 compiles recent, decision-relevant outcomes for soil hydraulic, chemical, and biological responses to biosurfactant use. Place this table after

the paragraph above and refer to it when selecting doses and delivery routes in later sections.

Biosurfactants also influence biological properties. In situ studies have documented shifts in microbial communities and enzyme activities that align with faster hydrocarbon turnover, suggesting a direct ecological effect beyond simple solubilization (Zhu et al., 2024). However, responses are dose-dependent. High rhamnolipid loads increased phytotoxicity of diesel-impacted soils and suppressed seed germination in multiple species; ryegrass experiments confirmed growth inhibition at elevated concentrations. These results underline the need for careful dosing and site-specific trials (D’Incau et al., 2024; Parus et al., 2023). Relative to a common synthetic benchmark, rhamnolipids generally exhibit lower plant toxicity, yet even green surfactants can impair growth if overapplied (Gidudu and Chirwa, 2022). Biosurfactant use alongside plant–microbe systems is therefore best framed as a calibrated intervention that raises bioavailability while preserving soil biota (Parus et al., 2023; Sonowal et al., 2021).

APPLICATIONS AND RECENT CASE STUDIES

Biosurfactants are now used in several practical soil clean-up routes. First, soil washing with rhamnolipids has been applied to weathered soils and drill cuttings. In one study, washing a petroleum-contaminated soil with about five hundred milligrams per liter rhamnolipid achieved 58.5% TPH reduction under optimized conditions, and

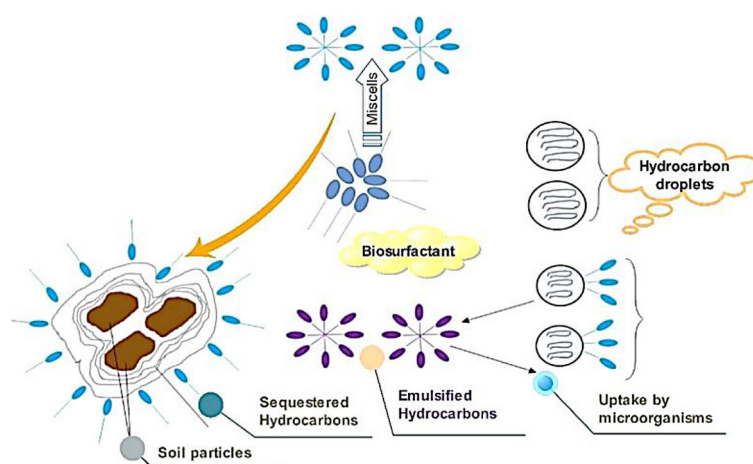


Figure 3. Mechanisms of biosurfactant action in soils, including hydrocarbon desorption from soil particles, micelle formation, emulsification, and enhanced uptake by microorganisms

Table 2. Selected effects of biosurfactants on soil properties

Soil property	Biosurfactant and context	Key outcome	Ref.
Hydraulic behavior	Surfactant-mediated wetting in hydrophobic porous media	Increased infiltration due to reduced surface and interfacial tension	(Ogunmokun & Wallach, 2023)
Chemical balance in saline soils	Rhamnolipid amendment, controlled studies	Soil organic matter increased 5–31.6% and electrical conductivity decreased about 0.3–42.4%	(Liu et al., 2023)
Field salinity near roots	Rhamnolipid drip irrigation in cotton	Rhizosphere desalination $\approx$ 9.7% (slight), 4.5% (moderate), 2.5% (high) salinity classes	(Chen et al., 2024)
Microbial community and enzymes	Biosurfactant use during remediation	Community structure and core enzyme activities shifted toward hydrocarbon degradation	(Zhu et al., 2024)
Plant response and safety window	Rhamnolipid in diesel-impacted soils	Higher phytotoxicity at 600 mg kg ⁻¹ ; seed germination depressed; microbial activity reduced	(Parus et al., 2023)
Plant response under high dose	Rhamnolipid on ryegrass	Growth inhibition and oxidative stress markers increased at high concentrations	(D'Incau et al., 2024)
Comparative toxicity	Rhamnolipid vs SDS (synthetic)	Rhamnolipid less phytotoxic than SDS while still inhibitory at high dose	(Gidudu & Chirwa, 2022)
Integrated soil–plant systems	Biosurfactant-enabled phyto-microbial remediation	Improved plant-assisted degradation and soil biological function	(Sonowal et al., 2021)

higher removal in drill cuttings, showing that biosurfactant washing can serve as an effective first stage before biological polishing (Olasanmi and Thring, 2020).

Second, bioaugmentation combined with a sophorolipid dose improved hydrocarbon removal and shortened half-life. Adding 1.5 g sophorolipid per kilogram to an indigenous consortium raised TPH removal from 12.2% to 57.7% and cut the degradation half-life from 32.5 days to 20.4 days (Feng et al., 2021). Third, hybrid materials enhance performance. Sophorolipid-modified biochar delivered 62.86% removal after sixty days, compared with 52.64% for unmodified biochar and 24.09% in the control, while enriching functional catabolic genes and petroleum-degrading taxa (Prakash et al., 2021). Fourth, bio-electrokinetic systems couple a low-voltage field with biosurfactants to move and degrade oil in low-permeability soils. A laboratory study reported about 92% removal with biosurfactant-assisted BIO-EK versus 60% for electrokinetics alone over two days, indicating strong synergy when mass transfer is limiting (Arpornpong et al., 2020).

Finally, sequential treatment is gaining traction. Field-relevant drill cuttings were treated by a bio-based washing agent followed by bioremediation, reducing TPH to 0.9% after 49 days and pointing to feasible on-site workflows with green surfactants (Fanaei et al., 2020). Table 3 summarizes decision-relevant outcomes from these studies to guide dose selection and process integration in practice.

PRACTICAL DOSING WINDOWS AND FIELD SAFETY INDICATORS

Field deployment of biosurfactants in petroleum-contaminated soils benefits from translating laboratory efficacy into operational windows that balance hydrocarbon mobilization with ecological tolerance. For soil washing/ex-situ flushing, rhamnolipids (RL) perform reliably in the 0.2–1.0 g·L⁻¹ band, with many studies converging near $\approx$ 0.5 g·L⁻¹ and contact times on the order of 30–60 min; an illustrative optimization reported 500 mg·L⁻¹, 30 min, and soil/liquid ratios between 1:1 and 4:1 as effective for PHC reduction in both drill cuttings and field soils (Olasanmi and Thring, 2019). Reviews of washing practice similarly report RL or sophorolipid (SL) liquors around $\sim$ 0.5 g·L⁻¹ in oily matrices, often adjusted to sample hydrophobicity and mixing energy. Where SLs are used as co-surfactants (e.g., RL/SL blends) or at elevated temperature, comparable concentration windows are typical, with formulation choice guided by CMC behavior and sludge rheology (Zhao et al., 2024).

For in-situ soil amendment supporting bio/phyto-remediation, dosing should start conservatively and be piloted in microcosms. A broad evidence synthesis shows RL concentrations in terrestrial systems spanning 75 mg to 15 g·kg⁻¹ soil, with notable phytotoxic effects frequently emerging in the $\sim$ 200–600 mg·kg⁻¹ band and strong species-dependence of plant response (Parus et al., 2023). In particular, $\approx$ 600 mg·kg⁻¹ RL

Table 3. Recent applications of biosurfactants for petroleum-contaminated soils

Application	Matrix	Biosurfactant system	Key outcome	Note	Ref.
Soil washing	Weathered soil	Rhamnolipid solution	58.5% TPH removal under optimized washing	Effective first step before bioremediation	(Olasanmi and Thring, 2020)
Bioaugmentation + biosurfactant	Contaminated soil	Sophorolipid with indigenous consortium	57.7% TPH removal; half-life down to 20.4 d	Improves desorption and microbial activity	(Feng et al., 2021)
Hybrid sorbent-biosurfactant	Agricultural soil	Sophorolipid-modified biochar	62.86% removal vs 52.64% biochar and 24.09% control	Strengthens bioavailability and catabolic genes	(Prakash et al., 2021)
Bio-electrokinetic	Loamy soil, lab scale	Bacterial biosurfactant with low-voltage field	~92% removal vs 60% EK alone in 2 d	Useful for tight, low-permeability zones	(Arpornpong et al., 2020)
Sequential washing + bioremediation	Drill cuttings	Bio-based washing agent with lipopeptide	TPH down to 0.9% after 49 d	Cost-aware on-site workflow	(Fanaei et al., 2020)

decreased seed germination across several terrestrial species in diesel-spiked soils, underscoring the need to avoid aggressive single-shot additions in vegetated plots (Marecik et al., 2012).

Pragmatically, practitioners initiate RL at tens to low-hundreds $\text{mg} \cdot \text{kg}^{-1}$ (often split-dosed) and only escalate within data-supported bounds when bioassays and enzyme readouts remain favorable (Eras-Muñoz et al., 2022; Li et al., 2018; Parus et al., 2023). For SLs, microcosm evidence indicates that $\sim 0.5\text{--}1.5 \text{ g} \cdot \text{kg}^{-1}$ can enhance TPH biodegradation under certain conditions; however, vegetated systems warrant sub- $\text{g} \cdot \text{kg}^{-1}$ starts and explicit phytotoxicity screening due to species-specific sensitivity (Hosseini et al., 2025). Hybrid carriers (e.g., SL-modified biochar) are increasingly used to temper free-phase exposure while sustaining bioavailability gains, improving biodegradation trajectories relative to biochar alone (Chen et al., 2024).

Where electrokinetic (EK) assistance is applied to tight or fine-grained matrices, injectates in the $\sim 0.5\text{--}2 \text{ g} \cdot \text{L}^{-1}$ range for RL/SL have been reported to improve polycyclic aromatic hydrocarbon mobilization, provided that pH and conductivity gradients are actively managed at electrodes; recent bioelectrokinetic reviews outline voltage management and buffer strategies to minimize collateral stress while retaining mass transfer gains (Gidudu and Chirwa, 2021).

Operational safety indicators should run in lockstep with efficacy metrics during bench-to-pilot scaling. The seed germination index (GI) remains a sensitive, inexpensive sentinel; as a rule of thumb, $\text{GI} \geq 80\%$ of uncontaminated reference is interpreted as non-phytotoxic, 50–80% suggests moderate inhibition warranting dose hold or formulation change, and $< 50\%$ indicates

high phytotoxicity and a need for immediate de-escalation or delivery redesign (Kong et al., 2023). Because biosurfactants can increase xenobiotic bioavailability, GI responses should be read jointly with microbial and enzymatic endpoints. Soil dehydrogenase activity (DHA) – a proxy for microbial respiratory capacity – responds sensitively to PHC stress and recovery and is widely recommended as a core bioindicator during remediation; declining DHA with dose escalation should trigger a return to the previous safe setting and/or adoption of sorbent-mediated delivery (Mekonnen et al., 2024). Routine pH and EC tracking is advised (biosurfactant solutions and mobilized ions can shift both), and earthworm acute toxicity (OECD 207) remains a simple ecological backstop in pilot soils prior to wide deployment (Gidudu and Chirwa, 2022; Narenkumar et al., 2024).

In practice, we recommend a stepwise workflow: (i) bench jar tests to locate CMC-adjacent doses and screen GI at 72 h; (ii) 28-day soil microcosms tracking TPH decay plus GI and DHA; (iii) plot-scale pilots with split dosing and sentinel bioassays; and (iv) bounded scale-up only when GI and DHA remain at or near reference and no adverse shifts are observed in pH/EC or earthworm survival. This workflow operationalizes biosurfactant use within explicit dosing windows while enforcing quantitative safety gates that are inexpensive to monitor yet ecologically meaningful.

CHALLENGES AND FUTURE PROSPECTS

Production and cost remain the main obstacles. Biosurfactant manufacture still faces limited

titers and productivities, while downstream separation raises unit costs above those of many synthetic surfactants; recent process reviews repeatedly emphasize low fermentation yield, foaming control issues, and costly purification as key constraints [61, 62]. Progress in engineering safe, non-pathogenic production hosts and in redesigning media and bioreactors is narrowing this gap, but a dependable industrial supply adequate for large remediation campaigns has not yet been secured (Miao et al., 2024).

Feedstocks and circularity provide a practical path to improved economics. The use of secondary substrates and agro-industrial residues can lower feedstock costs and enhance life-cycle indicators; studies that integrate such residues report high sophorolipid titers and favorable techno-economic outcomes, indicating that scale-up can proceed without direct competition with food resources (Carolin et al., 2022; Roelants et al., 2024). Formulation and delivery in soils are critical yet underreported. Field soils vary widely in pH, salinity, organic matter and mineralogy; these factors shift the critical micelle concentration and increase adsorption losses, which in turn alter the effective dosing required for remediation. Moreover, formulation stability and storage behavior are infrequently addressed in environmental applications, and standardized stability and performance tests are needed prior to field deployment (Ghisman et al., 2025).

Defining a robust safety window is essential because benefits are dose-dependent. Excessive biosurfactant concentrations can be detrimental – for example, high rhamnolipid loads have been reported to inhibit plant growth and stress seedlings – thus placing an upper bound on doses

used in revegetation and restoration efforts. Future practice should couple efficacy assays with rapid plant and soil-biota screens to establish site-specific safety margins (Crouzet et al., 2020).

Evidence at full field scale remains limited. Much of the literature is confined to laboratory or pilot studies; although field-ready workflows have been proposed, multi-site validations and standardized, regulator-friendly protocols are still scarce. Faster integration of biosurfactants into established remediation frameworks will accelerate regulatory acceptance and build end-user confidence (Sun et al., 2024).

Four practical directions emerge. First, strain and pathway engineering to produce safe chassis that deliver high titers with reduced foaming and simplified downstream handling (Zhou et al., 2023). Second, continuous bioprocessing combined with in-situ product removal (for example foam fractionation or tailored precipitation) to lower downstream separation costs (Pang et al., 2024; Wang et al., 2024).

Third, development of decision-driven formulations that match local soil chemistry while respecting ecological toxicity limits, deployed alongside hybrid remediation techniques such as bio-electrokinetics or biochar-assisted delivery (Parus et al., 2023). Fourth, design for scale through circular feedstocks and supply models that align remediation demand with biomanufacturing capacity (Lan et al., 2023).

Taken together, advances in safer and more productive strains, leaner processing routes, site-matched formulations, and circular supply strategies offer a realistic pathway to translate promising laboratory results into dependable field practice.

Table 4. Selected production and recovery advances with decision-relevant outcomes

Lever	System and study highlight	Quantitative outcome	Ref.
Waste-to-biosurfactant	Secondary feedstocks for sophorolipids	High titers with favorable techno-economics in fed-batch scenarios	(Miao et al., 2024)
High-efficiency producer	<i>Starmerella bombicola</i> sophorolipids	Titers and productivities reported at up to $>200 \text{ g L}^{-1}$ and $\approx 2 \text{ g L}^{-1} \text{ h}^{-1}$	(Roelants et al., 2024)
Two-stage fermentation	Rhamnolipids, staged process strategy	$\approx 55\%$ productivity increase vs single stage	(Zhou et al., 2023)
Engineered safer chassis	<i>Pseudomonas putida</i> rhamnolipids	Titer $\approx 19.8 \text{ g L}^{-1}$ with mixed carbon source in engineered strain	(Pang et al., 2024)
Directed strain improvement	Rhamnolipid mono-congener production	Titer 26 g L^{-1} at 48 h; 17.9% higher than parent	(Wang et al., 2024)
Foam fractionation recovery	Continuous fractionation of rhamnolipids	Enrichment $\approx 2.5\times$, recovery $\approx 51\%$; batch runs up to $\approx 81\%$ recovery reported	(Mahamad et al., 2025)
Advanced isolation step	Heat-assisted precipitation for RLs	Efficient isolation from broth when conventional separation is difficult	(Luo et al., 2024)

Table 4 lists production-side advances with quantitative outcomes that are directly relevant to cost and supply planning for soil remediation.

CONCLUSIONS

In summary, this review provides a soil-specific, mechanism-to-design synthesis for deploying biosurfactants in petroleum-impacted soils. It delivers a decision matrix that aligns biosurfactant class and dose with site chemistry and process objectives (soil washing versus in situ amendment), articulates CMC-normalized dosing and practical delivery rules (including split dosing), and situates biosurfactants within integrated remediation trains (bioaugmentation, phytoremediation, electrokinetics, sorbent-assisted strategies) under defined ecological safety windows monitored by sentinel bioindicators (germination index, dehydrogenase activity). By connecting molecular/solution behavior (head–tail chemistry, CMC, surface/interfacial-tension control, wetting, micelle/emulsion regimes) to quantifiable soil outcomes across heterogeneous conditions (texture, organic carbon, salinity, PHC weathering), the review addresses a persistent gap between mechanism and design choice. Future work should prioritize multi-site pilots with harmonized endpoints, standardized formulation/stability testing under representative chemistries, techno-economic and life-cycle benchmarking, and advances in safe, high-titer, circular manufacturing to secure supply for field deployment.

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