



A Review on Biosurfactants

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ABSTRACT

Biosurfactants are produced by different microorganisms like bacteria, algae, fungi on to the cell surface. They are amphipathic in nature which helps them to aggregate at the interfaces, thereby reducing the surface tension. They are characterized by the molecular weight into two i.e. low molecular and high molecular. According to the different biomolecule such as protein, carbohydrate moiety attached, they have distinct and different structures which provide them different functions and applications in different fields. In this present review, the relationship among microorganism, biosurfactant and hydrocarbon has been described and the different applications in various industries have also been explained.



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INTRODUCTION

Biosurfactants are surface active compounds mainly synthesized by microorganisms of different genera such as bacteria, microalgae, yeasts, fungi etc. [1] on cell surface or extracellularly or cell bounded such as lipopeptides or lipopolysaccharides [2]. Different biosurfactant producing bacteria include *Pseudomonas* sp., *Bacillus* sp., *Alcaligenes* sp. *Serratia* sp. [3]. Fungi that were reported to producing biosurfactants were *Candida lipolytica*, *Candida bombicola*, *Ustilago maydis*, and *Aspergillus ustus* [4]. The different yeasts are also found to be biosurfactant producers and they were *Torulopsis bombicola*, *Torulopsis petrophilum*, *Torulopsis apicola* and *Candida antartica* [5]. They are applied in bioremedi-

ation of water and large-scale production of crops, MEOR, environment clean-up, industrial effluent treatment, detoxification, heavy metal degradation, increasing shelf life, and pesticide bioremediation. In medicine field, they are used for gene transfection, immunoglobulins etc.

Structure and Types of Biosurfactant

Biosurfactants are amphipathic in nature that helps them to decrease the surface and interfacial tensions by aggregating at the interfaces of immiscible liquids like oil and water [1]. Since, biosurfactants have both hydrophobic and hydrophilic parts in their structure that assists them to aggregate between the interfacial phases of liquids of different polarities [6]. The hydrophobic or lipophilic part consists of protein or peptide with a chain of 10 to 18 carbon atoms and the hydrophilic or lipophobic moiety contains ester, phosphoric acid, carboxylic acid, sugar, or amino acid group [7]. Biosurfactants have been classified into two groups i.e. low and high molecular mass. The low molecular mass includes glycolipids, lipopeptides and phospholipids and high molecular mass consists of polymeric and particulate surfactants [8]. Biosurfactants have been named after the sugar, phosphate, or amino acid moiety that are present in their structures and divided into different groups which are shown in Figure 1.

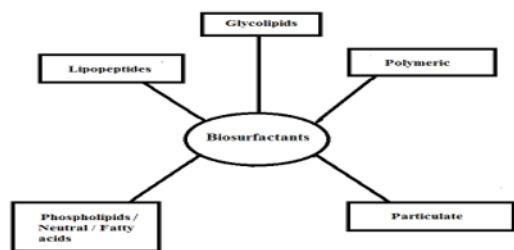


Figure 1: Different Groups of Biosurfactants

Glycolipids

Glycolipids are mono or disaccharide acylated with long fatty acid chain. Different classes of biosurfactants under glycolipids are shown in Figure 2. Rhamnolipids are mainly produced by *P. aeruginosa* but other than that *P. chlororaphis*, *Acinetobacter* sp., *Enterobacter* sp. have also been reported to have the capability to produce glycolipid containing biosurfactants. Trehalose lipids are mainly produced by species having mycolic acid associated with the cell wall [9].

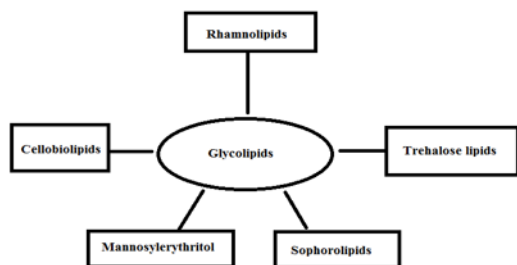


Figure 2: Types of Glycolipids

The reported microorganisms are *Mycobacterium* sp., *Nocardia erythropolis*, *Arthrobacter parrafineus*. Sophorolipids is made up of carbohydrate sophorose linked with fatty acid mainly produced by yeast species. *Torulopsis bombicola* and *Candida lipolytica* are known producers of sophorolipids [10]. Cellbiolipids are another type of glycolipids produced by *Ustilago zaeae*, and *Ustilago maydis* [1]. Mannosylerythritol are produced by *Pseudomyzma* sp. as major compound than *Ustilago* and *Pseudozyma siamensis* [11].

Lipopeptides

Lipopeptides are made up of peptides from amino acid origin which is linked to the lipid part of the fatty acids. The different classes of lipopeptides have been shown in Figure 3 [12]. Lipopeptides are mainly synthesized by *B. amyloliquefaciens*, *B. subtilis*, *B.licheniformis*, *B.clausii*, and *B. brevis*. Other than these *Pseudomonas* sp., *P.fluorescens*, *Psyringae*, and *Virgibacillus salarius* are also reported to be producers. The different

types of lipopeptides are fengycin, bacillomycin D, surfactin, lichenysin, iturin A, amphisin, gramicidin S, viscosin, and serrawettin and they are found to be produced by *Serratia marcescens* [13]. Three molecular types of serrawettin are produced by different enzymes such as W1 whose synthesis is controlled by serrawettin W1 synthase W2 production is controlled by serrawettin W2 synthase and W3 is controlled by serrawettin W3 synthase [14].

Phospholipids

Phospholipids are made up of two fatty acids esterified to the glycerol having a phosphate residue at the third position which are mainly produced by different *Bacillus* species.

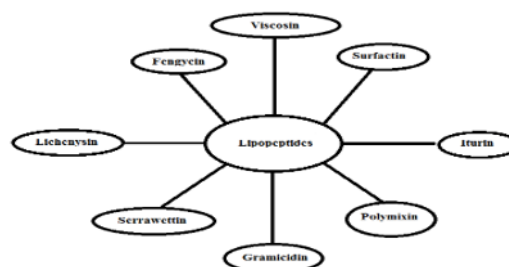


Figure 3: Types of Lipopeptides

Bacilosocin, is an antibiotic produced by *B. subtilis* is a phospholipid. Another *B. subtilis* produced phospholipid which could inhibit the growth of *E.coli*, *S. aureus* and *P. aeruginosa*. It was Lim et al reported *B. koreensis* producing different cellular phospholipids which were phosphatidylglycerol, diphosphatidyl glycerol, phosphatidylethanolamine and two unidentified phospholipids (PL1 and PL2). *B. gottheilii* was also found to produce phospholipid. The other phospholipid producing bacteria are *B. salsus*, *S. aureus*, *Corynebacterium urealyticum* [15]. Neutral lipids are synthesized by *Myroides*, *N. erythropolis*, *R. erythropolis* [16]. Different types of phospholipids are shown in Figure 4.

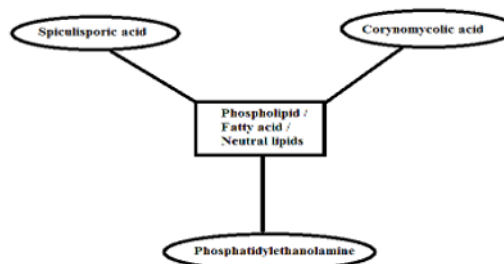


Figure 4: Types of Phospholipids

Polymeric Biosurfactants

Polymeric biosurfactants are high molecular weight biosurfactants and are generally emulsifiers. The different types of polymeric biosurfactants are listed

in Figure 5. Emulsan which could stabilize oil-water emulsions was isolated from *A. calcoaceticus*. Chamanrokh et al isolated *A. calcoaceticus* RAG-1 from Persian Type Culture Collection, IROST which was also found to produce emulsan [17].

Alasan, an anionic biosurfactant made up of alanine heteropolysaccharide was extracted from *A. radiore-sistens*. Alasan purified from *A. radiore-sistens* KA53 was named as apo-alasan. Liposan produced by *C. lipolytica* bioemulsifier and found to be made up of 83% heteropolysaccharide and 17% amino acids. *Yarrowia lipolytica* was also found to be able to produce liposan which stabilized oil-in-water emulsions [18].

Mannoprotein is an effective bioemulsifier was observed and purified from *S. cerevisiae*. *C. albicans* was reported to produce mannoprotein that elicited delayed hypersensitivity. Protein PA extracted from *S. aureus* could stimulate Interleukin alpha, Interleukin-4, Interleukin-6, Tumor necrosis factor and Interferon gamma. Protein PA i.e. Pediocin was found to be produced by methylotrophic *Pichia pas-toris* species. Biodispersan production by *A. cal-coaceticus* A2 at an optimum pH between 9 and 12 was observed by Rosenberg [19]. Secretion of biodispersan by *A. calcoaceticus* A2, an extracellular heteropolysaccharide was reported.

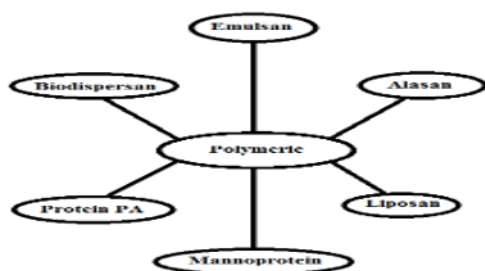


Figure 5: Types of Polymeric Biosurfactant

Particulate Biosurfactants

These particulate biosurfactants are also high molecular weight emulsifiers or biosurfactants. There are two types of biosurfactants in this group i.e. vesicles or whole microbial cell acts as biosurfactants. *A. baumannii* elicited immune response by the outer membrane vesicles. *A. baumannii* producing outer membrane vesicles inhibited the host growth was reported and can be used against *A. baumannii* infection. Fulsundar et al. [20] observed that antibiotic stress affected the process of vesiculation in *A. baylyi*. *Cyanobacteria* and *A. calcoaceticus* has the ability to be a biosurfactant i.e. whole microbial cell was found by Karanth et al. [21].

Source

Microorganisms are the sources of biosurfactants

and are ubiquitous in nature. The different bio-surfactant producing microorganisms are bacteria, fungi, yeast, archaea etc. Different microorganisms have been found to be isolated from diesel contaminated soils, industrial waste, vegetable waste, organic soil. Biosurfactant producing microorgan-isms were isolated from a wide range of envi-ronments and some of them were cited here. *S. marcescens* had been reported to be present in soil and freshwater samples. Sastoque-Cala et al isolated *Rhizobacteria* from fique [22]. Bhardwaj et al. iso-lated *Fusarium proliferatum* from oil contaminated sludge of rice bran [23]. Chaillan et al. reported aerobically growing fungal species *Amorphoteca*, *Neosartorya*, *Talaromyces*, *Graphium* that were iso-lated from petroleum contaminated soil [24]. Chail-lan et al. isolated *Candida*, *Yarrowia*, *Pichia* which were aerobically grown from petroleum contami-nated soil [24]. Halophilic archae such as *Natrono-coccus*, *Halorubrum* were isolated from Soda lakes, Egypt. Haloarchae was isolated from saline lake, Algeria. Saponins, lecithins, humic acids are phy-togenic based biosurfactants from plants and these surfactants are released from decaying roots of plants.

Relationship

The relationship among microorganism, biosurfactant and hydrocarbons was shown in Figure 6. Microorganism produces biosurfactant in the presence of water insoluble substrates like hydrocar-bons as biosurfactants have the ability to increase the transport of nutrients across the membrane. Biosurfactants help in the uptake of hydrocarbon as energy sources and microorganisms solubilise it with the help of biosurfactant [11]. Hydrocarbons act as energy sources for the microorganisms which are known hydrocarbon degraders and this can only happen in the presence of biosurfactant which split up and solubilise the hydrocarbon.

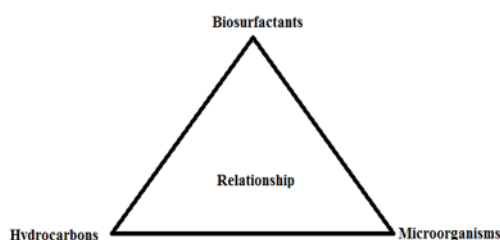


Figure 6: Relationship Among Microorganisms, Hydrocarbon and Biosurfactant

Properties of Biosurfactants

The different roles of biosurfactants are emulsifica-tion, dispersion, wetting, antimicrobial activity and

others are shown in Figure 7. Zhang et al. observed that rhamnolipid produced could solubilise and disperse phenanthrene [25]. This property helps their enlarged uptake of insoluble hydrophobic substrates through their cell membrane. The lipopeptide, surfactin produced by *B. subtilis* had antibiotic, anticlotting, haemolytic and antiviral properties. Bacilosocin, a phospholipid produced by *B. subtilis* was found to have antibiotic property. DPPH and superoxide scavenging ability was more than 50% against hydrogen peroxide by mannosylerythritol. Surfactin produced by *B. subtilis* exhibited good reducing power for DPPH and ferrous chelating ability. *B. polyfermenticus* KJS-2 producing lipopeptide also revealed similar antioxidant role biosurfactant.

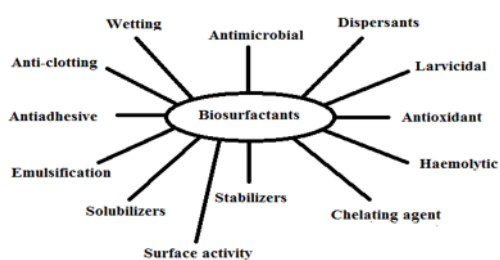


Figure 7: Properties of Biosurfactants

The stabilizing and high surface activity of biosurfactant help in the formation of microbubbles that assist in molecular imaging, sludge water treatment. These antioxidant, stabilizing roles of biosurfactants help them apply in food and cosmetic industries [26].

Biosurfactants produced by *C. albicans* showed antimicrobial and antiadhesive activities towards variety of urinary tract infectious pathogens. Pseudofactin II produced by *P. fluorescens* BD5 lowered adhesion of *E. coli*, *E. faecalis*, *E. hirae*, *S. epidermis*, *P. mirabilis* and *C. albicans*.

The wetting ability of biosurfactant enhances the pesticide bioavailability in soil and also helps in remediation of soil contaminated with polycyclic aromatic hydrocarbons. Sophorolipid produced by *Cryptococcus* sp. showed efficient removal of Zn (II) by its chelating activity than the synthetic surfactant.

Stenotrophomonas maltophilia NBS-11 reported 65% iron chelating effect after optimization of biosurfactant production. Restoration of contaminated soil from petrol by using rhamnolipids from *Withania somnifera* was found by Kumar et al. [27]. The different roles of these microbial compounds increase the use of biosurfactants in industrial applications as shown in Figure 8.

Applications

Environment

The removal of hydrophobic substrates that are tightly bound to the soil are mainly removed by desorption by biosurfactants. This type of property is very much significant for microorganisms which are involved in biodegradation and bioremediation process. Biosurfactant produced by *C. tropicalis* was reported to remove >95% of motor oil from soil by Batista et al. [28].

The cell bound biosurfactant produced by *Lactobacillus pentosus* could remediate the hydrocarbon contaminated soil [29]. Biosurfactant produced by *Pseudomonas* sp. proved having the ability to enhance the remediation of chemical insecticides such as chlorpyrifos and beta-cypermethrin from the agricultural soil. Bioremediation of pesticide and chlorinated hydrocarbons by surfactin and glycolipid have been reported, respectively. There have been reports of degradation of endosulfan sprayed on cashew plantation by biosurfactant produced by *Bacillus subtilis*.

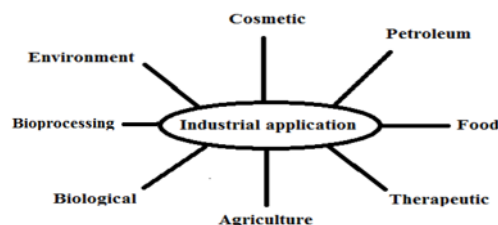


Figure 8: Different Industrial Applications of Biosurfactants

Biological

The different biological applications include motility, biofilm formation, quorum sensing, resistance towards toxic compounds, storage of carbon and energy molecules and a protective mechanism against high ionic strength [30]. Biosurfactants have been reported to have to increase adhesive properties in microbes to different surfaces and increase the colonization of various interfaces and thus shows antibiotic properties against other microbes. Surfactin also has been reported to help in the formation of fruiting body in *B. subtilis*. Surfactin production and flagellar synthesis are associated with swarming ability by *B. subtilis* was revealed by Kearns and Losick [31].

Petroleum Industry

In contrast to the synthetic surfactants, the advantages of biosurfactants can be employed in the exploration of new sites of heavy oil and in processing of petroleum i.e. extraction, transportation and

storage. Biosurfactants are also used in microbial enhanced oil recovery, oil transportation from one site to another through pipeline and also cleaning of oil contaminated vessels [32]. During the transportation of oil through pipelines, due to the viscosity there are chances of deposition of asphaltenes or paraffin and also drop in pressure. Biosurfactants involved in petroleum industry are by modifying wetting ability in reservoirs, reduction in viscosity of oil, dispersing oil, control in deposition of asphalt or paraffin, stabilization of oil emulsion etc. Biosurfactants improve in mobilizing oil and enhances the recovery of crude oil from reservoirs by microbial enhanced oil recovery [33].

Therapeutic

Biosurfactants being biologically active with a broad range of chemical structures give wide applicability than the synthetic medicines and antimicrobial agents due to their safe and effective therapeutical applications. Kitamoto et al. explained that mannosylerythritol lipids exhibited antimicrobial activity against gram-positive bacteria [34]. Biosurfactant produced by probiotic bacteria such as *L. lactis* and *S. thermophilus* showed antimicrobial activity against a variety bacterial and yeast strains were studied by Rodrigues et al. [35]. Mannosylerythritol lipids synthesized by *Candida* strains showed immunological and neurological properties grown on vegetable oil.

Food Industry

Increasing demands of the consumers for lowering the addition of artificial chemicals i.e. additives and ingredients, biosurfactants are been used as a natural synthesized one due to their different beneficial properties. Biosurfactants and their surface activity are not affected at high or low temperature, pH, salt concentrations. In industrial applications, stability at different temperature, salt concentration and pH conditions are necessary which are observed in the biosurfactant produced by different microorganisms like *B. licheniformis* JF-2, *Arthrobacter protophormiae*, *Bacillus subtilis*. Biosurfactant from *C. utilis* was used as emulsifying agent in salad formulations. Yeast cultivated on whey based medium was used as a potential food bioemulsifier [36]. Lipopeptide from *B. subtilis* showed stable emulsions with soyabean and coconut fat revealing its potentiality to use as emulsifying agent in food industry.

Agriculture

Productivity of farming land usually matters due to the presence of organic and inorganic hydrocarbon or pollutants present in the soil. So biosurfactants can be used effectively for remediation of

these hydrocarbons or contaminated soil [37]. For improving of soil quality soil washing and clean-up treatment are used [38]. Biosurfactants produced by microorganisms have antimicrobial activity that is considered a helping tool for biocontrol for good crops that encourage against parasitism, competition, systemic resistance and hypovirulence as reported by Singh et al. [39]. Jazzar and Hammad had found that biosurfactants have the ability to improve insecticidal activity [40]. Boyette et al. found that the biosurfactant in combination with fungus i.e. *Myrothecium verrucaria* could eradicate weeds that affected land productivity [41].

Advantages of Using Biosurfactants

Some usually possess antimicrobial activity like surfactin produced by *B. subtilis* have been reported to have antimicrobial activity [42]. Plipastatins, produced by *B. cereus*, from a family of lipopeptides was reported to have antifungal activity [43]. Their diverse chemical structures, biodegradability and ecological friendliness have increased their applications in industries like food, paper, agriculture, building construction, beverage, cosmetics, leather, pharmaceutical industries and oil bioremediation [44].

They are capable to be modified by genetic engineering and biotechnology and increase the solubility of insoluble form of hydrophobic hydrocarbons such as polycyclic aromatic hydrocarbon (PAH) [45]. They can be synthesized and produced from cheaper agro-based substrates, animal feedstock and waste materials from industries and thus helps in the cleaning process of polluted soil and environment [46]. Low toxicity compared to chemical treatment and good stability at extreme temperatures, pH and NaCl concentrations help us to be used in oil remediation, microbial enhanced oil recovery, cleaning up of oil contaminated pipes, remediation of soil and ground water [47].

CONCLUSION

This review provided information regarding the different applications of biosurfactants in various industries in the current scenario. Since biosurfactants are not yet in a position to be compared to the chemical surfactants because of its economic perspective. However, these biomolecules are promising solutions for many current and upcoming problems due to its wide applications.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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