Review of Biosurfactant – Producing strains of Bacillus subtilis

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***Abstract* - Biosurfactants are surface active compound that reduce the interfacial tension between two liquids or that between a liquid and solid. Biosurfactants special properties are nontoxic, easily biodegradable, eco-friendly and high stability and wide variety of industrial application makes them highly useful group of chemical compounds. Biosurfactant are produced from variety of microorganism. For the removal of heavy metals from industrial waste the biosurfactant as an active biological surface-active agent may provide an alternative solution. Biosurfactant show the properties of reducing surface and interfacial tension, stabilizing emulsions, promoting foaming, high selectivity, and specific activity at extreme temperature, pH and salinity, the ability to be synthesized from renewable resources.**

***Index Terms* - Properties of biosurfactant, classification, advantages and uses of biosurfactant.**

INTRODUCTION

Surfactants are compound that lower the surface tension between two liquids or between liquid and solid. Surfactant may act as a detergent, wetting agents, foaming agents and dispersants.

Microbial surfactant is the surface-active molecule derived from a large number of microorganisms. Surfactants are organic compound that contain both hydrophobic and hydrophilic moieties. Thus, surfactant contains both waters insoluble. i.e., water soluble group as well as water repellent group i.e., hydrophilic group. Biosurfactants are synthesized by microbes like bacteria, fungi, and yeast. Biosurfactant include the properties of dropping surface tension, stabilizing emulsions, promoting foaming and are generally nontoxic and biodegradable. Recently interest in biosurfactant has increased because of it is diversity, flexibility in operation and more eco-friendly than chemical surfactant. Contrary to the chemical surfactants that are generally produced from petroleum feed stock, the microbial surfactant can be produced by using a wide variety of cheap agro-based raw materials. The features that make them commercially superior to their chemically synthesized counterparts are their stability at extremes of temperature, pH and salinity. These properties are desirable in various industrial process such as in food processing, pharmaceutical formulation, and enhanced oil recovery and in environmental bioremediation.

Biosurfactant are found in the nature in a wide variety of chemical structures including glycolipids, lipopeptides and lipoproteins, fatty acids, neutral lipids, phospholipids, polymeric and particulate lipids. Significant consideration has been given in the past to the blend of surface-dynamic molecules from biological source because of potential use in food processing oil industry and p'cology. Even though the type of quantity of the microbial surfactants produced depends mainly on the producer organism, factors like nitrogen and carbon, temperature, air circulation and minor components additionally influence their creation by the organism.

Mineralization is governed by desorption of hydrocarbon from soil. Surfactant can raise the surface area of hydrophobic resources, such as pesticides in water and soil surroundings, thus increasing their water solubility. Later, the existence of surfactants might increase microbial degradation of pollutants. The utilization of biosurfactants for the degradation of pesticides in soil and water environment has gained significance recently.

The biosurfactants offers following benefits over them chemically synthesized counterparts.

1) Biodegradability: owing to low toxicity and simple chemical structure, the compounds do not

persist in the environment and are degraded easily preventing problems by way of accumulation.

2) Biocompatibility and digestibility: biological origin imparts them inherent characteristics of compatibility which allows their unabated usage in cosmetics, pharmaceutical and as functional food additives.

3) Availability of raw materials: biosurfactants can be produced from relatively cheap raw material available in abundance. The carbon source ranging from hydrocarbons, carbohydrate to lipids may be used separately or in combination with each other microbial production.

4) Acceptable production economics: where permitted by intended use, biosurfactant can be produced even from industrial waste and by product, which remains a promising area for bulk production.

5) Environmental control: processes for stabilization of industrial emulsion, control of oil-spills, biodegradation and detoxification of industrial effluents and in bioremediation of contaminated soil can be favoured with use of biosurfactant.

6) Specificity: the presence of specific functional groups imparts specificity in the action by the biosurfactant molecules. This property can be paramount importance in detoxification of specific pollutants, de-emulsification of industrial emulsions, development of specific cosmetic, specialized pharmaceutical and food application. The most significant properties of these microbial products are effectiveness at the extremes of temperature, pH and salinity. Biosurfactant possess the characteristic property of reducing the surface an interfacial tension using the same mechanisms as that for chemical surfactant.

CLASSIFICATION OF BIOSURFACTANTS

Unlike the chemically synthesized surfactants that are generally categorized on the basis of on the type of the polar group present, biosurfactant are in general classified chiefly by their chemical composition and microbial origin. Rosenberg and Ron, suggested, biosurfactants could be divided into low molecular mass molecule that efficiently lower surface and interfacial tension, and large molecular mass polymers, that are more efficient as emulsion stabilizing agents.

Biosurfactants with their microbial sources -

1) Surfactin: microbial origin by Bacteria
Bacillus subtilis (Arima et al.1968)-
Bacillus licheniformis F2.2 (Thaniyavarn et al.2003)
Bacillus subtilis ATCC 21332 (Nitschke and Pastore, 2003)
Bacillus subtilis LB5a (Nitschke and Pastore, 2006)
Bacillus subtilis MTCC 1427 and MTCC 2423 (Makkar and Cameotra, 1999)

2) Surfactant BL86: microbial origin by bacteria
Bacillus licheniformis 86-(Horowitz and Currie, 1990)

3) Halobacillin:
Marine Bacillus sp. (Trischmann et al.1994)

4) Isohalobacillin:
Bacillus sp. A1238 (Hasumi et al. 1995)

Classification based on molecular weight: -

[1] Low molecular-weight biosurfactants: -
These are compound lower the surface and interfacial tension at the air/water interface. The low molecular weight biosurfactants are generally glycolipids or lipopeptides. The best studied glycolipids are rhamnolipids, trehalolipids and sophorolipids which are disaccharides that are acylated with long chain fatty acids or hydroxy fatty acids.

[2] High molecular weight biosurfactant: -
These are usually referred to as bio emulsion. They are more effective in stabilizing oil in water emulsions. They are highly efficient emulsifier that work at low concentrations and exhibit considerable substrate specificity. Desai and Banat stated that in general, the structure of a biosurfactants includes a hydrophilic moiety consisting of amino acids and peptides anions or cations, mono-, di- or polysaccharides and hydrophobic moiety consisting of unsaturated and saturated or fatty acids derivatives.

Classification based on chemical composition: -

a)Glycolipids: -

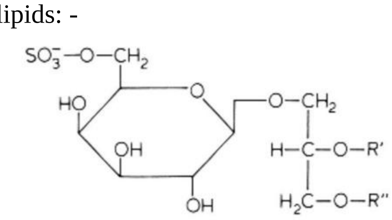

Fig 1

Mostly biosurfactants are glycolipids. They are lipids with a carbohydrate attached. The connection is by means of either an ether or ester group. Among the glycolipids, the best known are rhamnolipids sophorolipids and trehalolipids.

(b)Rhamnolipids: -

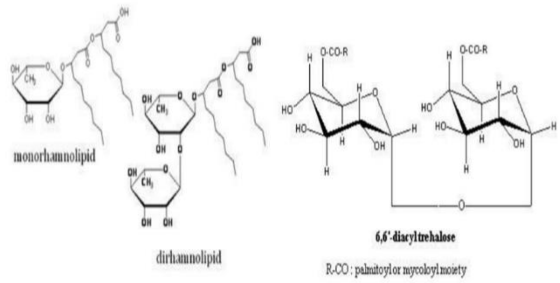

Fig.2 (a)Structure of mono and di rhamnolipid (b) Structure of Trehalose lipid

Rhamnolipids is a set of biosurfactant that studied extensively. These are formed by many species of Pseudomonas and have tremendous antimicrobial activity against several mutual microorganisms, which is an important property of all cosmetics due to the regular contamination of the product by the human touch. Rhamnolipids is a type of glycolipid biosurfactant that contain also one or two molecules of β-hydroxy decanoic acid. In 1999 the structure of rhamnolipid produced by P aeruginosa on mannitol and naphthalene by liquid chromatography was compared. And it was found that for mannitol the most common rhamnolipid contained two rhamnose and two 3-hydroxydecaonic acid and whereas rhamnolipid produced by naphthalene contained two rhamnose and one 3-hydroxydecaonic acid.

(c) Trehalolipids: Trehalolipids is the type of glycolipids that containing trehalose hydrophobic moieties. Such type of biosurfactant structure is various in hydrophobic moieties, varying from short fatty acid to long fatty acid chain. Overall, it is detected that the bacteria showing Gram's positive produces Trehalolipids biosurfactant.

(d) Sophorolipids: Sophorolipids glycolipids are produced by yeast these are the complex mixture of both free acid and lactone form. The acidic SL contain fatty acid as tail.SL lactonic are formed by the esterification between carboxylic end of the fatty acid and 4" of the sophorose end. Then the lactonic SL has better surface tension lowering property where as the acidic SL have better potential to form foam and solubility properties.

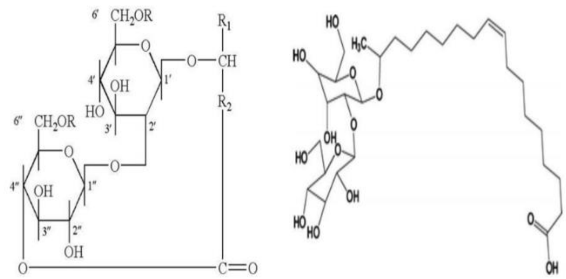

Fig.3. Structure of SL lactonized and acid Form.

Lipopeptides and lipoproteins:
Lipopeptide biosurfactants are cyclic compounds and they are generally isolated from Bacillus and Pseudomonas type bacteria. Lipopeptides mostly consist of hydrophilic peptides, mostly they consist 7 and 10 amino acids long, linked to a hydrophobic fatty acid structure. Bacillus cyclic lipopeptides contains three major groups known as the surfactin, iturin and fengycin families. Surfactin contains 7 amino acid cyclic sequences connected to a C13–C16 fatty acid.

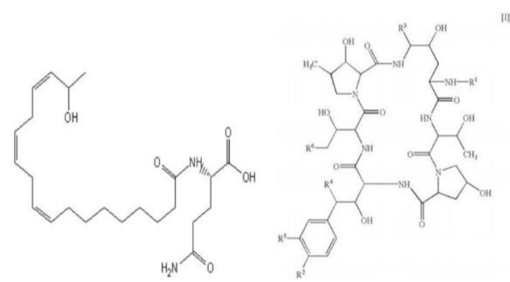

Fig.4 Structure of aminolipopetide and cyclic aminolipopetide

Polymeric Biosurfactant:
Polymeric biosurfactant are high weight molecular biopolymers which contain polysaccharides, proteins, lipopolysaccharides, lipoproteins or mixture of theses biopolymers. A variety of microbes produces polymer biosurfactant. High viscosity, tensile strength, and resistance to shear is the properties of polymeric biosurfactant. The example of different type of polymeric biosurfactant are, Ex: Acinetobacter calcoaceticus, Candida lipolytic.

Properties of biosurfactant: -
Biosurfactants are surface active compound. It gathers at the boundary between two immiscible fluids or between a fluid and a solid. By decreasing surface

(liquid-air) and interfacial (liquid-liquid) strain they diminish the repulsive forces between two distinct stages and permit them to mix and subsequently improve the solubility properties like chemical surfactant. Biosurfactant produces from B. subtilis is able to lower the surface tension of water to 25 Mn/m and interfacial tension of water/hexadecane to < 1 Mn/m.

In several cases it was found that biosurfactant activities are not influenced by environmental condition such as temperature and ph. In 1990 McInerney suggested that lichenysin formed by B. licheniformis was not affected by temperature (up to 50°C), pH (4.5– 9.0) and by sodium chloride and Calcium concentrations up to 50 and 25 g/l respectively. Aside from theses above properties biosurfactant can be easily degraded unlike the chemical surfactant and thus and they are they are predominantly appropriate for the natural applications like bioremediation, and scattering of oil spills. The toxicity of biosurfactants is much lower and some of the researcher consider as these is non-toxic compounds. Very few literatures are available that describes the toxicity of biosurfactant and their direct bad impact on environment. Therefore, they are suitable for pharmaceutical, food and cosmetic uses. A biosurfactant from P. aeruginosa was compared to a synthetic surfactant that is broadly used in the industry, regarding toxicity and mutagenic properties.

Advantages and uses of biosurfactant:

The unique properties of biosurfactant (Microbial Surface-Active Agents) such as low toxicity, relative ease of preparation and widespread applicability, make it different from chemical synthetic surfactant and present day it has become newly important product of biotechnology for industrial and medical applications and they allow to replacement of chemical synthetic surfactant. They can be used as emulsifiers, de-emulsifiers, wetting agents, spreading agents, foaming agents, functional food ingredients and detergents in many industrial sectors such as, Petroleum and Petrochemicals, Organic Chemicals, Foods and Beverages, Cosmetics and Pharmaceuticals, Mining and Metallurgy, Agrochemicals and Fertilizers, Environmental Control and Management, and many others.

There are many advantages of biosurfactant as compare to chemically synthesized surfactants like: -

1.Biodegradability: - Easy to biodegradable as compare to the chemical surfactant.

2. Low toxicity

3.Biocompatibility and digestibility, that permits their application in cosmetics, pharmaceuticals and food seasonings.

4.Easily availability of raw material: The raw material need for production of biosurfactant are easily available, biosurfactant producing microorganism can be isolated from the industrial waste like oil contaminated soil, petrol pump spilled, and also can be isolated from municipal waste.

5.Use in environmental control. Biosurfactants can be professionally used in handling industrial emulsions, control of oil spills, biodegradation and lowering the toxicity of industrial discharges and in bioremediation of contaminated soil.

6.Specificity in their action, since biosurfactant has specific organic functional group and frequently specific in their action. This is individually used in lowering the toxicity of the pollutant, used in increasing the emulsification property, used as raw material in cosmetic, medicinal and foodstuff applications.

CONCLUSION

From the review, it can be concluded that biosurfactant can be produced from verities of microorganisms. Bacterial strain has been isolated from various industrial wastes like agrowaste, coffee processing waste, petrochemical waste etc. It was found that the production of biosurfactant depends on bacterial growth. It was noted that the production of biosurfactant is still test on industry level because of high cost of bacterial nutrient, to overcome this problem, various researchers isolated bacteria from industrial waste as declared above. It also studied that the production rate of biosurfactant depends on bacterial growth, so the bacterial growth must be in optimal conditions like optimum temperature, and media ph.

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