



## Effects of rhamnolipids on microorganism characteristics and applications in composting: A review



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### ABSTRACT

Biosurfactant rhamnolipids have been applied in many fields, especially in environmental bioremediation. According to previous researches, many research groups have studied the influence of rhamnolipids on microorganism characteristics and/or its application in composting. In this review, the effects of rhamnolipids on the cell surface properties of microorganisms was discussed firstly, such as cell surface hydrophobicity (CSH), electrical, surface compounds, etc. Moreover, the deeper mechanisms were also discussed, such as the effects of rhamnolipids on the structural characteristics and functional characteristics of the cell membrane, and the effects of rhamnolipids on the related enzymes and genes. Additionally, the application of rhamnolipids in composting was discussed, which is an important way for pollutant biodegradation and resource reutilization. It is believed that rhamnolipids will play more and more important role in composting.

### 1. Introduction

With the development of the society, many kinds of industrial materials and products, such as petroleum (Ron and Rosenberg, 2014), pesticides (Zeng et al., 2013), medical waste (Duan et al., 2008), have caused a great pollution to water, soil and air. They seriously threaten the sustainable functioning of ecosystems and the human health (Liu et al., 2013). Hence, many efforts in development of environmental and eco-friendly chemicals and/or techniques should be carried out to strengthen pollution prevention and treatment.

Compared to harsh chemical and physical treatments, bioremediation has the potential to eliminate contaminants through biochemical mineralization in a permanently and cost-effectively way (Huang et al., 2008; Kumar et al., 2011). Additionally, bioremediation holds a variety of advantages over chemical and physical remediation, e.g., low cost, few or no by-products, reusability, eco-friendliness (Sun et al., 2016), and is widely applied in the pollution remediation. However, some pollutants (e.g., Polycyclic aromatic hydrocarbons (PAHs), Phenols, Petroleum hydrocarbons, Heavy metal (Liu et al., 2010; Liu et al., 2016)) possess high toxicity and low bioavailability to microorganisms, resulting in low efficiency and even failure of bioremediation (Zhong et al., 2016a). Therefore, some additives such as surfactants have been

used to improve the remediation efficiency in the application of practical bioremediation. These surfactants enhance the removal of contaminants may through increasing the apparent solubility and bioavailability of contaminants (Tang et al., 2014; Liu et al., 2017), changing the microbial surface properties more hydrophilic or hydrophobic and strengthening the interaction between pollutants and microorganisms (Liu et al., 2014b). Meanwhile, these surfactants also can increase the activity and/or quantity of related enzymes and genes in microorganisms (Liang et al., 2010) and reduce the toxicity of pollutants to microorganisms (Liu et al., 2010), which is also in favor of bioremediation of pollutants.

Commonly used surfactants are divided into synthetic surfactants and biosurfactants. They are amphiphilic molecules containing polar and non-polar groups and possess surface active properties (Banat et al., 2010), such as emulsification, dispersion, foaming, detergency, wetting and stabilization etc. (Gudiña et al., 2015; dos Santos et al., 2016; Zhong et al., 2016b). Biosurfactants are usually produced by microorganisms and have several advantages compared to synthetic surfactants, e.g., biodegradability, high environmental compatibility, strong surface activity and lower toxicity (Zhou et al., 2011b; Yu et al., 2015). Rhamnolipids are the most extensively studied biosurfactants for bioremediation of pollutants (An et al., 2011), especially for the

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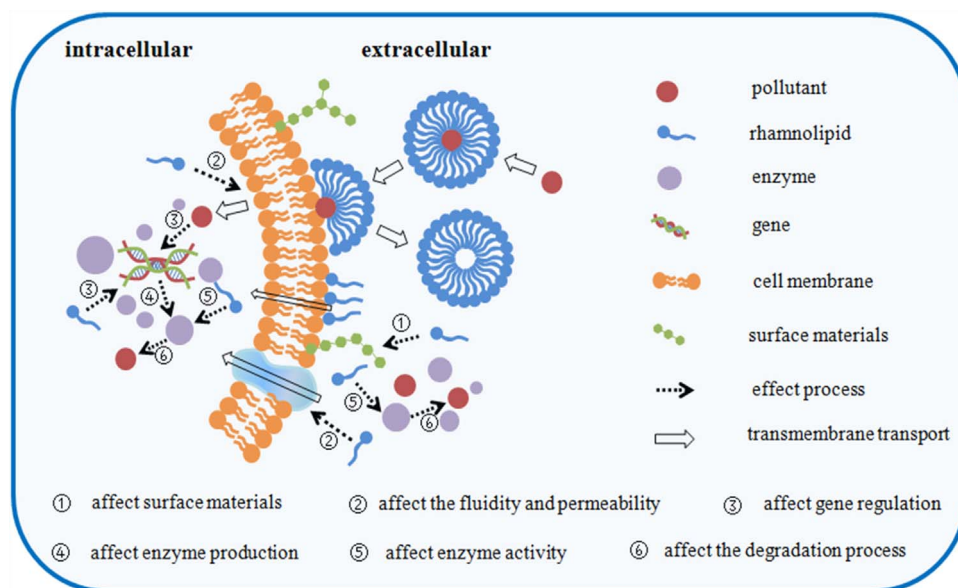


Fig. 1. The process of rhamnolipids effects on microorganism characteristics and the degradation of pollutants.

hydrophobic organic pollutants (Trellu et al., 2016).

It is well-known that the elimination of pollutants by microorganism is a systemic bioprocess and can be separated as two phases: a rapid removal process by adsorption and a following degradation process (Sun et al., 2016; Zhong et al., 2016c). The adsorption process is not only influenced by the types and physicochemical properties of pollutants, but is related closely to surface properties and cell membrane structure of microorganism (Luo et al., 2003). The degradation process is related to internal activities of microbes, and is catalyzed by degrading enzymes which is controlled by related genes. The bioremoval process including adsorption and degradation processes may experience alteration in the presence of rhamnolipids which exert impacts on the cell surface properties (e.g., CSH, cell surface charge, surface compounds, surface free energy (Fig. 1 ①)) and membrane structure (Fig. 1 ②) (Kaczorek et al., 2008; Zeng et al., 2011) and the internal activities (e.g., degrading enzymes (Fig. 1 ③–⑤)) of microorganism (Zeng et al., 2006).

This review mainly focuses on the impacts of rhamnolipids on the cell surface properties and the internal activities of microorganism. Studying the effects of rhamnolipids on the metabolic process may be an important way to illustrate the influence of rhamnolipids on bioremediation process, such as the metabolic pathways, intermediate product, metabolic dynamics, degrading enzyme activity and gene expression, etc. However, there are few studies about the effects of rhamnolipids on microbial metabolism, and there is less research about the effects of rhamnolipids on microbial degradation of pollutants on the genetic level. Meanwhile, the application of rhamnolipids in composting (an effective bioremediation technology) has also been discussed. The compost medium contains a variety of pollutants, and they can be effectively removed by microbial degradation (Kästner and Miltner, 2016). Studying the effects of rhamnolipids on microorganism characteristics is important to illustrate the mechanism of composting in the presence of rhamnolipids, and benefit to develop more effective bioremediation technology. The process of rhamnolipids effecting on microorganism characteristics and the degradation of pollutants is shown in Fig. 1.

## 2. Production and characteristic of rhamnolipids

Most of biosurfactants are produced by microorganisms. Based on their biochemical nature, biosurfactants can be classified into low-molecular and large-molecular compounds (Lovaglio et al., 2015). The

low-molecular biosurfactants, such as lipopeptides (e.g. surfactin and fengycins) (de França et al., 2015), glycolipids (e.g. rhamnolipids and sophorolipids) (Smyth et al., 2010) and phospholipids (e.g. phosphatidylethanolamine) (Janek et al., 2013), can efficiently lower surface and interfacial tension. The large-molecular biosurfactants, such as lipoproteins, lipopolysaccharide (LPS), proteins, polysaccharides and biopolymer complexes (e.g. emulsan and alasan) are more effective as emulsion stabilizing agents (Abdel-Mawgoud et al., 2010; Lovaglio et al., 2015).

Rhamnolipids are the most widely studied glycolipids biosurfactants. The discovery of them can be traced back to 1946 (Rikalovic et al., 2015). They are produced by a variety of species of microorganisms (e.g. bacteria, fungi, yeast), and the main producing species is the gram-negative strains of *Pseudomonas aeruginosa* isolated from various habitats (water, soil or even plants) (dos Santos et al., 2016) (Table 1). Other species, e.g., *Nocardioopsis* spp. (Roy et al., 2014), *Acinetobacter calcoaceticus*. (Rooney et al., 2009), *Enterobacter* spp. (Hoskova et al., 2013) and *Burkholderia* spp. (Hošková et al., 2015), also have good productions of rhamnolipids under the suitable conditions.

The production cost of rhamnolipids is high since the expensive raw materials used in bacterial fermentation and the complex purification process, that limits the application of rhamnolipids in industrial scale (Henkel et al., 2012). Therefore, many efforts have been carried out in order to reduce the costs and increase yield in the former studies, such as using cheap substrates (Moya Ramirez et al., 2015), optimizing the production conditions (Borges et al., 2015), using different production process (Nalini and Parthasarathi, 2014), screening new natural producing strains (Roy et al., 2014), using genetic engineering strategy (Lovaglio et al., 2015), researching more efficient methods for separation and purification of rhamnolipids homologue (Zhang et al., 2016a) (Fig. 2). A lot of researches have evaluated low-cost substrates, developed efficient fermentation processes and screened new strains to increase the yields of rhamnolipids. However, little studies have been described in the genetic handling of strains to increase production efficiency (Lovaglio et al., 2015). Several important gene regulation process and key genes are involved in the complexity genetic network in the rhamnolipids production by *Pseudomonas aeruginosa* (Dobler et al., 2016). The biosynthesis of rhamnolipids includes three major steps, some key genes and quorum sensing signals: The synthesis of two important precursors during the first two steps, namely  $\beta$ -hydroxyalkanoyl- $\beta$ -hydroxyalkanoyl-ACP (HAA-ACP) and dTDP-L-rhamnose. Then, two special rhamnosyltransferases sequentially catalyze the condensation of the precursors to mono-rhamnolipids (mono-RL) and di-rham-

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