



## Sophorolipids: A review on production and perspectives of application in agriculture

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### Abstract

Sophorolipids are bioactive molecules that have gained a lot of attention in the recent decades due to their unique functional properties of reducing surface and interfacial tension, emulsification and solubilization. They are mainly produced by the yeast *Candida bombicola* and are composed of a sugar moiety linked to a fatty acid chain. Sophorolipids are non-toxic, highly efficient and stable at extreme conditions and possess environmentally friendly characteristics over the chemical surfactants. This review is focused on the main characteristics of sophorolipids, fermentation processes, and their utilization in the agricultural field. In this context, sophorolipids are very suitable for use in agriculture, as enhancers of solubility and mobility of plant nutrients, which could result in increased plant biomass, root size and fruit yield. In addition, they could be used for biodegradation of oils, bioremediation of heavy metals in contaminated soils, and as potential biopesticides, to control phytopathogenic microorganisms in agriculture. The extensive use of chemical pesticides has led to widespread insecticide resistance and to hazards to human health and the environment due to their high toxicity. Thus, the introduction of a new biomolecule to control plant diseases and increase crop yield has become an interesting alternative. As a result of the demonstrated antimicrobial activity towards phytopathogenic bacteria and fungi, sophorolipids could be extensively explored in the agriculture field, as a sustainable and natural multifunctional agent for plant crops and soils.

**Additional keywords:** antimicrobial; anti-phytopathogenic; bioremediation; fermentation; soil; biosurfactant; *Candida bombicola*.

**Abbreviations used:** FL (fiducial limits); IRM (integrated resistance management); MRR (resistant parent); MRS (F<sub>1</sub> offspring); RR (resistant allele); RS (offsprings); XF (log LC<sub>50</sub> of reciprocal crosses); XRR (bifenthrin-sel population); XSS (susceptible population).

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## Introduction

According to the Food and Agriculture Organization of the United Nations (FAO, 2019), the world population will increase from 6 billion people to 9 billion by 2050. Consequently, an increase in food consumption and agricultural production is expected (Oliveira *et al.*, 2015; Mishra & Arora, 2018). Therefore, the agriculture and biotechnology fields must ally to maintain productivity and environmental preservation. In the formulation of agrochemicals,

the addition of chemical surfactants is often used to confer emulsifying and dispersing properties. However, these compounds are considered pollutants, being easily accumulated in the soil and leached into groundwater (Morillo & Villaverde, 2017). An alternative could be the substitution of chemical surfactants for biosurfactants. These molecules are produced by microorganisms and possess low toxicity and high biodegradability (Olanya *et al.*, 2018).

Sophorolipids are biosurfactants synthesized in high concentrations and generally by non-pathogenic stra-

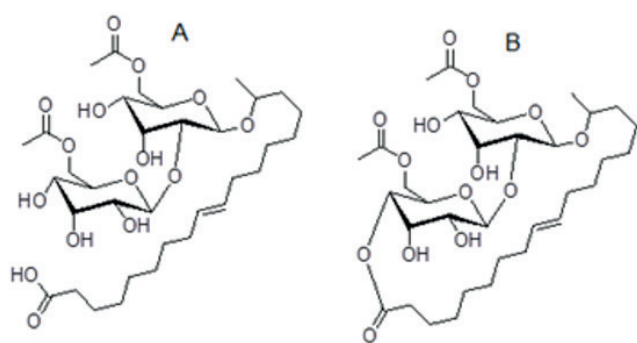
ins, making this group of molecules particularly attractive for commercial productions and future applications considering safety aspects (Van Bogaert *et al.*, 2007; Paulino *et al.*, 2016). They are currently applied as biosurfactants in the industry and their products are commercially available (Sharma & Oberoi, 2017).

In addition to their emulsifying properties, sophorolipids present potent antimicrobial activity and can be used as partial or total substitutes for pesticides, germicides and synthetic sanitizers (Giessler-Blank *et al.*, 2012; Kosaric & Vandar-Sukan, 2014; Olanya *et al.*, 2018). Additionally, they can also be utilized in the removal of heavy metals, hydrocarbons and antibiotics from contaminated soils, facilitating the biodegradation of these contaminants by reducing surface and interfacial tensions (Ahueke *et al.*, 2016; Minucelli *et al.*, 2017; Sun *et al.*, 2018).

This review presents the main characteristics of sophorolipids, showing the important parameters in the production using conventional substrates and agro-industrial residues. The application of sophorolipids in pesticides, germicides and sanitizers formulations, as well as their utilization in bioremediation of contaminated soils, is also considered.

### Sophorolipids: structure, microorganisms and production conditions

Sophorolipids are glycolipids structurally composed of a sophorose disaccharide (2'-O- $\beta$ -D-glucopyranosyl-1- $\beta$ -D-glucose) linked by a  $\beta$ -glycosidic bond to a long-chain of fatty acids (Jiménez-Peñalver *et al.*, 2016; Solaiman *et al.*, 2017). They are produced as a mixture of chemical structures, in which the carboxylic end of this fatty acid is either free, presenting an acidic or open form, or internally esterified at the C4', C6' or C6" position, resulting in the lactonic form (Fig. 1). These structures may also differ in regarding to the number of carbons, unsaturation, hydrogenations and acetylation. This chemical structural variation depends on the substrates and parameters used in the fermentation process, reflecting in the physical-chemical



**Figure 1.** Chemical structure of A) acidic and B) lactonic sophorolipids

aspects and in the biological properties of the sophorolipids (Diaz de Rienzo *et al.*, 2015; Jadhav *et al.*, 2019). In general, acidic sophorolipids are more soluble and have a better foaming capacity, whereas the lactonic have better surface tension and antimicrobial properties (Zhang QQ *et al.*, 2017; Zhang X *et al.*, 2017).

*Candida apicola* and *C. bombicola*, described in 1961 and 1970, respectively, were the first microorganisms used to produce sophorolipids (Claus & Van Bogaert, 2017). Nowadays, several sophorolipids producers have been reported (Table 1).

*C. bombicola* (*Starmerella bombicola*) is the main microorganism used for sophorolipids production, due to its high yields up to 400 g/L (Pekin *et al.*, 2005). This yeast is considered safe for human health and possess the Generally Recognized as Safe status (GRAS) (Van Bogaert *et al.*, 2011; Shah *et al.*, 2017). *C. bombicola* was isolated from environments with high osmotic pressure, such as honey, nectar and pollen. Then, the production of sophorolipids may be related to the adaptation of yeasts to the high concentrations of sugars in their habitat (Hommel *et al.*, 1994; Van Bogaert *et al.*, 2007).

Sophorolipids biosynthesis initiates with lipases, that promote the releasing of fatty acids, which may undergo to  $\beta$ -oxidation or hydroxylation, resulting in a hydroxylated fatty acid chain. Hereafter, two molecules of UDP-glucose will be added through the enzymes glycosyltransferase I and II, that couple two molecules of glucose at the C1 and C2 positions to the hydroxyl group  $\omega$  or  $\omega-1$  of the fatty acid, forming the non-acetylated acidic sophorolipids (Asmer *et al.*, 1988; Van Bogaert *et al.*, 2011). The enzymes responsible for the lactonic and acetylated structures are lactonesterase and acetyltransferase, respectively (Bajaj *et al.*, 2012).

This biosurfactant is considered a secondary metabolite, being produced at the end of the exponential phase and in the beginning of the stationary phase of the fermentation. They are synthesized from a single hydrophilic carbon source (carbohydrate) or by association with hydrophobic sources (lipids, hydrocarbons, vegetable oils or animal fat). Studies have shown that the production is increased when both sources are available (Cooper & Paddock, 1984; Asmer *et al.*, 1988; Davila *et al.*, 1994). The hydrophilic substrate is directed to produce the sophorose portion, while the hydrophobic component is the source for the lipid tail.

Glucose and oleic acid are considered the preferred sources of the metabolism for sophorolipids production (Asmer *et al.*, 1988; Bajaj *et al.*, 2012; Kosaric & Vandar-Sukan, 2014; Jadhav *et al.*, 2019), although the utilization of other sources has already been reported, as well as the use of agro-industrial residues to lower the production costs.

Several hydrophobic sources have been tested in the production of sophorolipids, and an increase in the yield

**Table 1.** Microorganisms producing sophorolipids

Microorganisms	References
<i>Candida albicans</i>	Kurtzman <i>et al.</i> , 2010
<i>Candida apicola</i>	Kurtzman <i>et al.</i> , 2010
<i>Candida floricola</i>	Van Bogaert <i>et al.</i> , 2011
<i>Candida kuoi</i>	Price <i>et al.</i> , 2012
<i>Candida riiodocensis</i>	Kurtzman <i>et al.</i> , 2010
<i>Candida stellata</i>	Kurtzman <i>et al.</i> , 2010
<i>Candida tropicalis</i> Y9	Chandran & Das, 2011
<i>Candida parapsilosis</i>	Garg <i>et al.</i> , 2018
<i>Cryptococcus</i> sp	Van Bogaert <i>et al.</i> , 2011
<i>Starmerella bombicola</i>	Chen <i>et al.</i> , 2019
<i>Rhodotorula bogoriensis</i>	Van Bogaert <i>et al.</i> , 2011
<i>Rhodotorula babjevae</i> YS3	Sen <i>et al.</i> , 2017
<i>Candida bombicola</i> ATCC 22214	Dolman <i>et al.</i> , 2017; Jiménez-Peñalver <i>et al.</i> , 2018
<i>Wickerhamiella domercqiae</i>	Ma <i>et al.</i> , 2014
<i>Wickerhamomyces (Pichia) anomalus</i>	Van Bogaert <i>et al.</i> , 2011

was observed according to an increase in the number of carbon atoms in the fatty acids chain (C18 > C16 > C14 > C12) (Cavalero & Cooper, 2003). The palmitic (C16) and stearic (C18) acids are directly incorporated, while longer fatty acid chains are less bioavailable. Short fatty acid chains need an elongation by the addition of carbons or are easily metabolized via  $\beta$ -oxidation and are not incorporated due to biochemical restrictions of cytochrome P450 monooxygenase CYP52M1, which results in low sophorolipids production (Davila *et al.*, 1994; Konishi *et al.*, 2018).

In addition, many organic and inorganic nitrogen sources were tested, such as urea, peptone, NaNO<sub>3</sub>, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, malt extract and yeast extract (Ribeiro *et al.*, 2013; Jiménez-Peñalver *et al.*, 2018; Chen *et al.*, 2019). Yeast extract is considered the most efficient source as it also contains other nutrients and salts, such as pantothenic acid, thiamine and pyridoxine, and traces of zinc, iron and magnesium. At low concentration, sophorolipids production is stimulated, although, at high levels, it can stimulate the primary metabolism and deplete the source of glucose. Yeast extract also interferes in the balance of acidic and lactonic forms. In general, low concentrations of yeast extract associated with long fermentation time promote the increase of lactonic forms (Casas & Garcia-Ochoa, 1999).

Oxygenation is also a very important parameter in the fermentation process. During the exponential phase, yeasts are sensitive to oxygen limitation (Guilmanov *et al.*, 2002), and during the synthesis of sophorolipids, the enzyme P450 monooxygenase involved in biosynthesis, requires molecular oxygen (Van Bogaert *et al.*, 2011).

The optimum pH for *C. bombicola* growth is between 5.0 to 6.0 (initial pH) and 3.5 during the stationary phase to produce sophorolipids (Davila *et al.*, 1997). Usually, the pH decrease during this phase is due to the consumption of the nitrogen source and the generation of organic acids. Daverey & Pakshirajan (2009) report that

pH control at 3.5 can promote up to 27.6% increase in sophorolipids production. According to the literature, the optimum temperature to sophorolipids production varies from 25 to 30°C (Felse *et al.*, 2007; Kim *et al.*, 2009; Dolman *et al.*, 2017).

### Sophorolipids: Biosynthesis using agro-industrial wastes

Biosurfactants can be produced from conventional sources, such as glucose and oleic acid, or alternative substrates, for instance agro-industrial residues (Samad *et al.*, 2017). The raw material used in the production of sophorolipids corresponds to 10-30 % of the total costs (Gudiña *et al.*, 2015). Therefore, the use of low-cost substrates may be an alternative to turn the production process more viable economically, while also contributes to decrease environmental impacts (Daverey & Pakshirajan, 2009; Banat *et al.*, 2014; Satpute *et al.*, 2017).

Several alternative substrates for the production of sophorolipids have been already studied: animal fat, fatty acid residues, used cooking oil, dairy waste water, deproteinized whey, soy molasses, by-products of biodiesel and of soybean oil refining, wheat and rice straw, manioc flour, sugar beet and cane molasses, maize meal, potato, soybean and corn husk and sugarcane bagasse (Banat *et al.*, 2014; Santos *et al.*, 2017). Some pretreatments are needed to improve the production, for example sugarcane molasses must be clarified before use for removal of inhibitory components, and corn husks needs to be hydrolyzed to facilitate absorption and increase final yield.

Minucelli *et al.* (2017) studied the production of sophorolipids by *C. bombicola* with molasses and sugarcane juice, sucrose, glucose, poultry industry residual fat and sunflower oil. The production was optimized by statistical methodologies, and reached 39.81 g/L in the medium

containing 75 g/L of residual fat from the poultry industry, 77.5 g/L of glucose, 2.5 g/L of yeast extract, at 30 °C, 150 rpm for 120 h, demonstrating the possibility of applying agro-industrial residue for a good production of sophorolipids.

In other study, Hoa *et al.* (2017) produced sophorolipids of *C. bombicola* in sugarcane molasses and coconut oil (10%), using a temperature of 25 °C, pH 6, 180 rpm for 168 h. The authors obtained a maximum yield of 10 g/L. Therefore, it is important to emphasize that substrates from renewable sources are well regarded to substitute the first-generation substrates, although they must present adequate nutritional value for microbial growth as well as a considerable and competitive production and yield (Satpute *et al.*, 2017). Table 2 shows residues and/or by-products used to produce sophorolipids.

### Sophorolipids and their applications in agriculture

In agricultural practices, plants can be attacked by phytopathogens, mainly fungi and bacteria, at various stages of their development, from planting to storage stages, generating large economic losses. This problem was greatly minimized after the emergence of synthetic pesticides, which became popular in controlling pathogens, due to their immediate effectiveness and easy of application (Oliveira *et al.*, 2015; Mishra & Arora, 2018).

Chemical surfactants present in pesticides act as emulsifiers, dispersants and wetting agents, promoting increased product efficiency (Rostas & Blassmann, 2009). However, these surfactants, for example alcohol ethoxylates (AEOs), alkylamine ethoxylates (ANEOs), polyethoxylated tallowamines (POEAs), organosilicon and trisiloxane (Krogh *et al.*, 2003; Vaughn *et al.*, 2014; Mesnage & Antoniou, 2018), are considered organic pollutants (Petrovic & Barcelo, 2004) due to their accumulation in the soil, causing damage to the environment and human health (Blackwell, 2000; Morillo & Villaverde, 2017). Considering the adverse effect of synthetic

pesticides, several alternatives are being explored. Biosurfactants have attracted the attention of researchers because they are considered environmentally safe and can be used not only as adjuvants, but also as active compounds (Kosaric & Vandar-Sukan, 2014).

The antimicrobial action of sophorolipids occurs through their amphiphilic nature, which promotes synergistic interactions between the sophorose and the fatty acid portions and produces the surfactant effect, reducing interfacial and surface tension of the compounds, leading to destabilization and alteration of the membrane permeability of the pathogen (Seung-Hak *et al.*, 2003; Lahkar *et al.*, 2015; Valotteau *et al.*, 2017). These effects depend on the proportions of sophorolipids structural forms, since lactonic forms have better antimicrobial activity than acidic ones. The producer microorganism and the type of pathogen can also deeply influence the antimicrobial activity (Sharma *et al.*, 2016; Zhang QQ *et al.*, 2017), which has been more effective against gram-positive bacteria than against gram-negative and fungi (Ribeiro *et al.*, 2013).

Some reports demonstrated the sophorolipids antimicrobial activity *in vitro*. A study conducted by Lang *et al.* (1989) showed the inhibitory effect of sophorolipids on the growth of the conidia germination of *Glomerella cingulata*, a phytopathogen typically found in apples, grapes and other fruits. Yoo *et al.* (2005) presented the sophorolipids as an antimicrobial agent for the control of *Botrytis cineria*, commonly known as gray mold and infects diverse vegetables, for example strawberry, grape and tomato. Sen *et al.* (2017) also tested the inhibitory effect of sophorolipids produced by *Rhodotorula babjevae* YS3 against *Colletotrichum gloeosporioides*, *Fusarium verticillioides*, *Fusarium oxysporum*, *Corynespora cassiicola* and *Trichophyton rubrum*, demonstrating their promising application against these plant pathogenic microorganisms.

Giessler-Blank *et al.* (2012) reported that sophorolipids may improve the activity of pesticides and herbicides. In their work, they used as study plants barley variety 'Ingrid' and blue grass (*Poa pratense*). Sophorolipids were tested as a curative and protective agent, and as an adjuvant to the agrochemicals epoxiconazole, sulfur and

**Table 2.** Wastes and by-products to produce sophorolipids

Substrates	Production (g/L)	References
Beet molasses and residual oil cake (petroleum)	25.10 g/100 g of substrate (SSF)	Jiménez-Peñalver <i>et al.</i> , 2018
Hydrolyzed corn straw and used oil	52.10	Samad <i>et al.</i> , 2017
Sugar cane molasses	10.00	Hoa <i>et al.</i> , 2017
Chicken fat and sugar cane molasses	39.81	Minucelli <i>et al.</i> , 2017
Rice bran	56.00	Ahueke <i>et al.</i> , 2016
Rice straw	53.70	Liu <i>et al.</i> , 2016
Corn stalk hydrolysate	43.80	Konishi <i>et al.</i> , 2015
Deproteinized whey	23.29	Daverey & Pakshirajan, 2010
Dairy industry wastewater and soy molasses	38.76	Daverey <i>et al.</i> , 2009
Sugar cane molasses	23.25	Daverey & Pakshirajan, 2009
Soy molasses	75.00	Solaiman <i>et al.</i> , 2007

rimulfuron. The phytopathogen used for the infection was the fungus *Blumeria graminis f. sp. hordei* (race A6). In their results, they observed that the association of pesticides with sophorolipids resulted in a synergistic interaction, increasing the efficacy of the pesticide alone from 46% to 99%. For the herbicide, there was also an increase in effectiveness from 53% to 72%, compared to the herbicide individually and combined with sophorolipids, respectively.

Schofield *et al.* (2013) studied the administration of pure, natural and modified sophorolipids as an antimicrobial agent *in vitro* and *in vivo*. In the first test, they observed the antimicrobial activity against diverse plant fungal pathogens (*Alternaria tomatophila*, *Alternaria solani*, *Alternaria alternata*, *Aspergillus niger*, *Aureobasidium pullulans*, *Botrytis cinerea*, *Chaetomium globosum*, *Fusarium asiaticum*, *Fusarium austroamericana*, *Fusarium cerealis*, *Fusarium graminearum*, *Fusarium oxysporum*, *Penicillium chrysogenum*, *Penicillium digitatum*, *Penicillium funiculosum*, *Phytophthora infestans*, *Phytophthora capsici* and *Ustilago maydis*) and plant bacterial pathogens (*Acidovorax carotovorum*, *Erwinia amylovora*, *Pseudomonas cichorii*, *Pseudomonas syringae*, *Pectobacterium carotovorum*, *Ralstonia solanacearum* and *Xanthomonas campestris*). In the *in vivo* tests in plants, the same authors verified the sophorolipids action against fungal infections, as well as their spores and zoospores. The plant species used in the tests were grapes, wheat variety 'Baart', rice and spinach. Modified sophorolipids showed an increase in antimicrobial activity compared to natural sophorolipids. According to the authors, the activity of the modified sophorolipids can be raised up to a thousand times in comparison with the natural sophorolipids, using simple modifications, such as esterification of carboxyl groups of fatty acids and selective acetylation of groups.

The germicidal effect of this biosurfactant was also explored in fruits and vegetables by Dengle-Pulate *et al.* (2014). The authors used sophorolipids produced in glucose and lauric alcohol as an active ingredient in a sanitizing solution containing  $\text{Na}_2\text{SO}_3$  and  $\text{Na}_2\text{CO}_3$ . The vegetables, chikoos (sapodilha), tomatoes, cucumber and lemons, were sprayed with the solution and the results showed total inhibition of *Erwinia chrysanthemi* ATCC 11663 and *Xanthomonas campestris* ATCC 13951 after 10 minutes of treatment. Olanya *et al.* (2018) also presented antimicrobial results *in vitro* and in tomato (post-harvest) against *Listeria monocytogenes*, *Salmonella enterica* and *Escherichia coli*. The results showed that the sophorolipids were effective at a concentration of 50 mg/L.

Additionally, sophorolipids were studied as adjuvants of herbicides and in the postemergence herbicidal activity. The plants used were the non-transgenic corn (*Zea mays* L. 'Silver Queen') and the sicklepod (*Senna obtusifolia* (L.) H.S. Irwin & Barneby), a weed commonly found in the United States. Vaughn *et al.* (2014) tested the ability of

sophorolipids to form emulsion with the herbicide lemon-grass oil. This is an important property, because lipophilic herbicides require an emulsifying agent to prevent separation into distinct phases. Moreover, the ability of a surfactant to enhance herbicide penetration is partially due to the amphiphilic nature. Normally, petroleum-based surfactants are the most widely used, although sophorolipids could form stable emulsions, representing a potential substitute for the synthetic surfactants.

Sieverding (2015) described in his work that sophorolipids were capable to increase the yield of agricultural crops. Tests were performed in the field, greenhouse and at laboratory scale on barley, wheat, soy and tomato (in the seeds and in the aerial parts of the plants). Plant infection was not induced, but occurred naturally. The application of sophorolipids was performed individually and in combination with fungicides, in order to observe the increase of total or partial biomass, related to the length of the roots, shoots or fruits. Surprisingly, it was found that sophorolipids were able to enhance the yield of agricultural crops, increasing 6.9 dt/ha compared to fungicide alone, which increased only 4.9 dt/ha.

The antimicrobial activity of sophorolipids is widely described in the literature against various types of bacteria and fungi. Despite the great potential of sophorolipids applications in the agriculture field, papers describing their action are still rare, though this action needs to be much more explored.

### Sophorolipids as a green alternative for soil bioremediation

Soil contamination is a major problem in the current environmental status and constitutes a barrier to sustainable development. It is mostly caused by inappropriate disposal of industrial discharge and mining tailings such as solvents, explosives, polycyclic aromatic hydrocarbons (PAHs), and polychlorinated biphenyls (PCBs) (Tabak *et al.*, 2005; Mao *et al.*, 2015). Heavy metals, such as lead, cadmium, chromium, copper, arsenic, zinc, mercury and nickel are also very common components in soils of industrialized countries (Mao *et al.*, 2015; Das *et al.*, 2016).

Several techniques for remediation have been developed over the years. Bioremediation has been pointed out as a good method, consisting in the use of living beings, or their components, in the recovery of contaminated sites (Corbu & Csutak, 2018). It has been highlighted as effective, easily available, ecofriendly and cost-effective in comparison with other approaches (Das *et al.*, 2016; Wijesekara *et al.*, 2017).

Biosurfactants have emerged as a promising alternative for bioremediation, because they facilitate the biodegradation of contaminants, while promotes its self-degradation and also increase the activity of microorganisms to

decompose the contaminants (Makkar *et al.*, 2011; Mao *et al.*, 2015; Shekhar *et al.*, 2015). The mechanism involved in bioremediation by biosurfactants is mainly based on their ability to form complexes with metals (micelles), thus increasing metal solubility and bioavailability in the soil solution (Seneviratne *et al.*, 2017).

Sophorolipids have gained a lot of attention in this field of application, being involved in solubilization and emulsification of hydrophobic contaminants (diesel, kerosene, crude oil, engine oil and motor oil), removal of heavy metals (chromium, lead, zinc, copper, cadmium, iron, mercury) and pesticides in aqueous phases (Ahueke *et al.*, 2016; Tang *et al.*, 2018).

Mulligan *et al.* (1999) tested the efficacy of different surfactants in contaminated soils, employing single and consecutive washings. Sophorolipids demonstrated the best results among the tested surfactants. In comparison with the treatment with 0.7 % HCl, its combination with 4 % sophorolipids presented an additive interaction and improved the removal of zinc and copper by 6 % and 18 %, respectively, after a single washing. The use of consecutive washings demonstrated even better results, removing nearly 100 % of both metals.

Another study tested the use of three surfactants to enhance biodegradation of hexachlorocyclohexane (HCH) isomers by *Sphingomonas* sp. NM05, obtaining that the introduction of rhamnolipids, sophorolipids and trehalolipids led to the increase of 30–50 % biodegradation. Also, among the surfactants tested, sophorolipids offered the highest solubilization and degradation, whereas stimulated the growth of bacteria biomass (Manickam *et al.*, 2012).

A soil contaminated with hydrocarbons and crude oil was used to evaluate the effectiveness of sophorolipids as a washing agent. It was noted that 30 % of 2-methylnaphthalene was effectively solubilized with 10 g/L of sophorolipids, a similar or even higher efficiency than that obtained with commercial surfactants. The crude oil was effectively removed by the addition of sophorolipids, resulting in 80% biodegradation in 8 weeks (Kang *et al.*, 2009).

Minucelli *et al.* (2017) utilized sophorolipids produced by *C. bombicola* in soil bioremediation for removal of lubricating-oil. The addition of sophorolipids enhanced significantly the CO<sub>2</sub> production in the first 4 days of incubation, indicating their fast effect. Also, sophorolipids showed a synergic interaction with *Bacillus subtilis* and *Bacillus licheniformis*, increasing microbial activity.

Other problematic type of contaminated soils is the application of livestock manure containing antibiotics, which impacts the natural selection on the soil microbiome (Peng *et al.*, 2015). Sophorolipids were used as a soil washing for contaminated site with cadmium, tetracycline, sulfadiazine, roxithromycin and antibiotic resistance genes. After washing with 20 g/L of sophorolipids solution, it was possible to remove almost all the contaminants. They also analyzed a lettuce cultivation after the treatment with

sophorolipids in comparison with the initial polluted soil, obtaining that physicochemical properties such as fresh/dry weight, root activity, chlorophyll and protein content significantly increased (Ye *et al.*, 2016).

A similar research conducted by Sun *et al.* (2018) focused on the use of sophorolipids to stimulate the dissipation of antibiotic resistance genes, tetracycline and microplastics of a contaminated soil. Results demonstrated that sophorolipids were good candidates, being able to break the inhibiting barrier of microplastics, enhancing the attenuation of the antibiotic and its resistance genes in the soil.

## Conclusion and future research directions

Due to their low toxicity, biodegradability and emulsification capacities, sophorolipids display many potential applications in diverse areas. The present review approaches the main properties and production parameters of sophorolipids, and their several potential applications in the field of agriculture, as adjuvants in pesticides and herbicides, germicide and their ant phytopathogenic activity. As pointed out, sophorolipids possess great antimicrobial activity against various types of bacteria and fungi, although their application *in vivo* against plant pathogens is lacking in the literature. The use of this biosurfactant in soil bioremediation can also open a more sustainable alternative for removal of contaminants such as heavy-metals, hydrocarbons, oils and antibiotics.

Even though their promising application has been demonstrated in many researches, some attempts must be done in order to make the application of sophorolipids viable. One main point is the optimization of production, since the chemical analogues surfactants have extremely competitive prices. The utilization of low-cost substrates is a primary alternative, exploring this eco-friendly appealing. Another attempt could be the use of genetic editing tools or further investigation of the metabolic pathways of the producer microorganism, aiming to improve and better understand the important variables in the sophorolipids production.

Further research is needed in the development of real scale application projects, either as exploring the ant phytopathogenic activity *in vivo* or for bioremediation in contaminated sites. Therefore, it can be concluded that sophorolipids have great potential and capacity to be explored in the agriculture field and further researches must be carried out.

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