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Research Article

Potential application of *Bacillus subtilis* SPB1 biosurfactants in laundry detergent formulations: Compatibility study with detergent ingredients and washing performance

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Surfactants play a very important role in laundry and household cleaning products ingredients. In this research, the application of lipopeptide biosurfactants, produced by *Bacillus subtilis* SPB1, in the formulation of a washing powder was investigated. The SPB1 biosurfactant was mixed with sodium tripolyphosphate as a builder and sodium sulfate as filler. The efficiency of the formulated detergent composition with different washing conditions to remove a stain from cotton fabric was examined. The results showed that the formulated detergent was effective in oil removal, with optimal washing conditions of pH, temperature, striate and time of washing system of 7, 65°C, 1000 RPM and 60 min, respectively. A comparative study of different detergent compositions (biosurfactant-based detergent, combined biosurfactant-commercial detergent, and a commercial detergent) for the removal of oil and tea stains, proved that the bio-scouring was more effective (>75%) in terms of the stain removal than the commercial powders (<60%). Moreover, the results demonstrated that the biosurfactant acts additively with a commercial detergent and enhances their performance from 33 to 45% in removing oil stain and from 57 to 64% in removing tea stain. As a conclusion, in addition to the low toxicity and the high biodegradability of the microbial biosurfactants, the results of this study have shown that the future use of this lipopeptide biosurfactant as laundry detergent additive is highly promising.

Keywords: *Bacillus subtilis* / Biosurfactant / Detergent / Formulation

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1 Introduction

For several years now, detergent products such as household, personal-care products, and industrial detergents, have depicted a constantly growing market [1]. Laundry detergent has traditionally been a powdered or granular solid, but the use of liquid laundry detergents has gradually increased over the years, and nowadays uses of liquid detergent equals or even exceeds the use of solid detergent. Some brands, also, manufacture laundry soap

in tablets and dissolvable packets [2]. Laundry detergents are complex mixtures of several active ingredients, up to 25 compounds [1], differing on the purpose of the detergent [3]. The major applications of detergent compositions are to clean fabric, crockery, cooking utensils, and hard surfaces such as glass, glazed surfaces, plastics, metals, and enamels [4]. The main ingredients of all laundry detergents are surfactants, builders, and fillers. Supplementary materials such as enzymes, dispersing agents, bleaching agents, and other auxiliary additives are also available in commercial formulations such as fabric softening clay, dye-transfer inhibiting ingredient and optical brighteners [1, 3, 5, 6]. Among these constituents, surfactants represent one of the most important laundry detergent ingredients [1, 6–9], comprising from 15% to 50% of the total detergent formulation [1, 6, 9]. As noted by Özdemir and Malayoglu (2004) [10], the properties and efficacy of detergent formulations are critically

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Abbreviations: SDS, Sodium dodecyl sulfate; SS, Sodium sulfate; STPP, Sodium tripolyphosphate

dependent on the interfacial activity of various surfactants, which are present in their composition. Being amphiphilic in nature, surfactants tend to partition preferentially at the interface between different phases (e.g. air/water, water/stain, stain/fabric) and reduce interfacial tension. In fact, the low surface tension of the water makes it easier to lift oil which can be the basis of cleaning dirt and grease off dirty dishes, clothes and other surfaces, and help to keep that oily dirt or grease suspended in the water thus forming emulsions. The hydrophilic head remains in the water and pulls the oil toward the water. Therefore, surfactants can work in three different ways: (a) they reduce the oil/solution and fabric/solution interfacial tensions and in this way, lift the stain of the fabric (Roll-up mechanism), (b) they reduce the oil solution interfacial tension and makes easy emulsification of the oil (Emulsification) and (c) through interaction with the micelles of a surfactant in a solvent (water), a substance spontaneously dissolves to form a stable and clear solution (Solubilization) [11].

Laundry detergents may contain more than one kind of surfactant [1,2,6]. According to their polar head group, surfactants are divided into four groups: anionic, nonionic, cationic, and amphoteric [2,5]. These surfactants differ in their ability to remove certain types of oil, in their effectiveness on different fabrics and in their response to water hardness [1,2,6].

Among surfactants, the anionic ones have the greatest wetting, foaming and emulsifying properties as compared with the cationic or non-ionic surfactants or biosurfactants [12]. In addition, Mishra (2009) [11] reported that the anionic surfactants are particularly effective in oily cleaning and oil/clay suspension. Still, they can react in the wash water with the positively charged water hardness ions (calcium and magnesium), which can lead to partial deactivation.

As described by Turbekar et al., (2014) [13], surfactants are most commonly derived from petrochemical sources. These chemically synthesized molecules are difficult to break down through the action of microorganisms [14], exert toxicity to fresh water living organisms and potentially cause environmental and toxicology problem due to their recalcitrant and persistent nature [8]. On the other hand, several authors had mentioned that one of the most challenging problems related to the use of chemically-based surfactants in cosmetic formulations, is their potential to cause skin irritations and allergic reactions [12]. Hence, their accumulation in the environment imposes adverse effects on natural resources and fabric treated with harsh chemicals is unsafe for human health [15]. In recent years, concerns regarding the environment on the part of consumers and new environmental control legislation have led to the development of natural surfactants as an alternative to existing products [1,9,14–16]. Moreover, the scientific community is motivated to seek surfactants that are more environmentally friendly, such as those achieved through microbial production, known as biosurfactants [8,14,17]. These biomolecules are used in the same way as the chemically synthesized surfactants. Likewise, they can be employed in detergency, emulsification, de-emulsification, wetting, foaming, dispersion, solubilization, among other possible activities or roles [1,8,12,19]. The wetting power allows the detergent solution to penetrate the pores until contamination with the impurities, the emulsifying activity maintains the impurities in solution, the foaming action causes the soiling,

the dispersing power prevents the aggregation of soiling and the solubilizing property dissolves organic liquids [20]. However, biosurfactants have advantages over their chemical counterpart some of which are structural diversity and stability at relatively high temperature and in adverse environments with the possibility of being produced from cheap raw materials [8,18,21–23]. They have also been found to possess several properties of therapeutic and biomedical importance: they have antibacterial, antifungal and antiviral properties and they have anti-adhesive action against several pathogenic microorganisms etc [22,24,25]. More importantly, biosurfactants have drawn the interest of different industries due to their higher biodegradability, low toxicity and biocompatibility [14,18,22,23,26]. The fact that they can be readily biodegraded means that they are significantly less damaging to the environment than the more recalcitrant chemical surfactants [12]. Therefore, thanks to their advantages, research has shown promising results of using biosurfactant in detergent formulation [5,9]. To cite, Kesting et al., (1996) [27] investigated the potential application of microbial surfactants in textile washing processes to remove various lipophilic preparations from fiber surfaces. Moreover, Bafghi and Fazaelpoor (2012) [5] investigated the use of rhamnolipids in the formulation of a washing powder. The results showed that the biosurfactant was effective in removal oil, chocolate and albumen stains from cotton. More recently, fabric wash analysis revealed that the crude glycolipids biosurfactants from *Pseudozyma* sp. NII 08165 removed stains efficiently and can be used in laundry detergent formulations [22]. In addition, the efficiency of a *Jatropha* oil derived sophorolipids biosurfactant produced by *C. bombicola* (ATCC22214) as a stain cleaner was reported by Joshi-Navare et al., (2013) [28]. Other studies reported that biosurfactants are used as detergents in cleaning up hydrocarbon/crude oil storage tank [18,19] and in a dish-washing product [29]. Moreover, the efficiency of the wetting behavior of biologically produced rhamnolipids respectively on glass, PET, polyvinylchloride (PVC), polyε-caprolactone (PCL) and a polymer blend PVC–PCL [30] and on glass, PET and gold surface [10] were evaluated. Nowadays, some biosurfactants based cleaners are used by some multinational companies. For instance, the US company SyntheZyme™ is developing bio-cleaners using modified sophorolipids as washing and cleaning agents (in liquid and powder form). Sophorolipids are also found in cleaning soap mixtures and all-purpose cleaners (such as interior cleaning spray, a window spray, a heavy-duty power cleaner, a car wash, and wax cleaner) produced by Ecover™. A leading detergent manufacturer, Henkel, started using sophorolipid surfactant in some of their regional branded glass-cleaning products, such as Sidolin, Instantanet, Sonasol, Tenn and Breff, all of which were sold in Europe. Indeed, the American company Naturell® has developed and patented a product for carpet cleaning containing enzymes and biosurfactants derived from the fermentation of sea kelp. The French company HTS BIO has created the product line Ecoway® for professional cleaning and maintenance that includes the cleaning product Biosurf [12]. In India, Akshay Intensive Marketing supplies Acticlean, a product containing non-specified biosurfactants, whose functions are claimed to be the improvement of wetting and spreading, detergency activity, modification and control of foam, emulsification, solubilization, and dispersion [31].

Previous study showed that a *Bacillus subtilis* strain SPB1 (HQ392822), isolated from a Tunisian soil contaminated with hydrocarbons [32], is an efficient producer of lipopeptide biosurfactant. As described by Mnif et al., (2016) [33], the mass spectroscopic analysis of the culture filtrates of SPB1 indicated the presence of surfactin, iturin and fengycin isoforms. These lipopeptide biosurfactants were found to be very effective in reducing surface tension of water from 70 mN/m to 34 mN/m with a critical micellar concentration (CMC) of 150 mg/L, indicating their high emulsifying property. Hence, it represents a suitable property in detergent industry. As suggested by Ghribi and Chaabouni (2011) [35], the SPB1 biosurfactant preserves its activity at extreme conditions of pH and temperature which is an extremely interesting feature in view of its potential use in detergent industries. Moreover, Mnif et al., (2013) reported that the SPB1 lipopeptide was used to enhance diesel solubility and mobility [36] and to improve textile dye biotreatment [37]. The in vivo potential toxicity of SPB1 lipopeptide biosurfactant towards male mice was performed by Sahounoun et al., (2014) [38]. They proved that it had no significant adverse effect on hematological parameters and serum biochemical data for a daily intake of doses lower than 47.5 mg/kg of body weight.

Owing these great interest and wide spectrum of application, the actual data was focused in evaluating the washing performance of the SPB1 biosurfactant in removing motor oil and tea from fabric. The detergent formulation was characterized for various chemo-physical properties and its effectiveness against commercial detergent was evaluated.

2 Materials and methods

2.1 Microorganism strain

The microorganism used through this work was a wild type strain isolated from the Tunisian soil and was identified as *Bacillus subtilis* SPB1 (HQ392822), by morphological, biochemical and 16S Ribosomal deoxyribonucleic acid (rDNA) sequence analysis [32].

2.2 Production and extraction of the crude biosurfactant

Previous study reported the culture conditions of producing biosurfactant from *Bacillus subtilis* SPB1 [35]. At the end of the cultivation, the bacterial cells were removed from surfactant contained in culture broth by centrifugation at 10000 RPM at 4°C for 20 min and the acellular culture broth served for biosurfactant extraction. The obtained supernatant was precipitated overnight at 4°C by adding concentrated HCl to achieve a final pH of 2. Gray white pellets formed by precipitation were collected by centrifugation at 10 000 RPM for 20 min, washed three times with acid water (pH 2) and dissolved in distilled water. The pH was adjusted to 8 with NaOH 1N and lyophilized. This serves as crude lipopeptide preparation to perform this study [39].

2.3 Washing material

Biosurfactant was obtained from *Bacillus subtilis* SPB1 at the National School of Engineering of Sfax (Tunisia). Analytical grade sodium tripolyphosphate (STPP) and sodium sulfate (SS) were used as builder and filler, respectively. One available commercially detergent on the Tunisian market and the chemical surfactant Sodium Dodecyl Sulfate (SDS) were used for a comparative study. A clean white cotton cloth cut into 2 × 2 cm pieces was stained firstly with 0.25 mL motor oil and secondly with 0.25 mL tea.

2.4 Washing procedure

As a first step, two sets of formulations were used in order to determine the best biosurfactant-based detergent formulation for oil removing. They were carried out at constant value of temperature, pH, striate and washing duration of the order of 20°C, 7, 750 RPM and 20 min, respectively. The first set, containing constant biosurfactant weight percentage of 10 and variable percentages of the filler and builder (Table 1), was used to determine the effect of builder in removing motor oil [5]. For the second set, efficiency of biosurfactant was studied by varying its percentages while keeping a constant builder weight percentage (Table 2) [5].

Table 1. Composition of the first set of formulation and effect of weight percentage of sodium triphosphate on the removal of oil

Surfactant (%)	Sodium triphosphate (%)	Sodium sulfate (%)	Oil removal in the presence of SPB1biosurfactant (%)	Oil removal in the presence of SDS (%)
10	0	90	33 ± 4.09	35.9 ± 4.94
10	20	70	32 ± 4.56	37 ± 5.99
10	50	40	33 ± 6.22	37.5 ± 6.16
10	60	30	35 ± 2.89	38.6 ± 4.35
10	70	20	38 ± 6.66	39 ± 9.16
10	80	10	38 ± 3.21	40 ± 8.09

pH = 7; temperature = 25°C; stirrer velocity = 750 RPM; washing time = 20 min.

(Data are expressed as mean ±SD for oil removal of duplicates).

Table 2. Composition of the second set of formulations and effect of weight percentage of SPB1 biosurfactant on the removal of oil

Surfactant (%)	Sodium triphosphate (%)	Sodium sulfate (%)	Oil removal in the presence of SPB1biosurfactant (%)	Oil removal in the presence of SDS (%)
0	70	30	24 ± 4.83	24 ± 1.22
5	70	25	32 ± 1.11	27.2 ± 4.73
10	70	20	38 ± 2.88	39 ± 9.16
15	70	15	36 ± 4.21	37 ± 6.63
20	70	10	35 ± 8.1	37 ± 8.42

pH = 7; temperature = 25°C; stirrer velocity = 750 RPM; washing time = 20 min.

(Data are expressed as mean ±SD for oil removal of duplicates).

The second step was realized to define the best washing conditions. In fact, the stained clothes were subjected to wash analysis under different conditions of pH, temperature, stirrer rate, and washing duration. The same experiments were carried out with detergent formulated with SDS instead of biosurfactant (positive control) and with detergent exempt of surfactant (negative control).

After fixing the best detergent composition and the best washing conditions for oil removing, a comparison study between the wetting properties of the biosurfactant-based detergent, the SDS-based detergent, the commercial detergent and a mixture of commercial detergent (90%) with biosurfactant (10%) [5] was carried out. Water was used as negative control. The same experiments were performed to evaluate the wetting properties of the tested formulation in removing tea from fabric.

The removal percentage of each stain was calculated using the precise weights of the pieces before and after washing [5]. In fact, each stained piece was kept for drying overnight at 25°C, washed in a flask containing 50 mL water with 2% detergent solution, rinsed twice in 100 mL distilled water and then dried.

3 Results and discussion

Thanks to their structural diversity, surface-active agents are able to reduce the surface tension of water significantly and then help hydrophobic materials to disperse easily in water which causes dirt and stain removal from the fabrics [11, 22]. In this study, the biosurfactant produced by SPB1 was applied as biosurfacting agent and the process was investigated for intended output parameters.

3.1 Optimization of detergent formulation on removing oil from cotton fabric

3.1.1 Evaluation of the effect of sodium tripolyphosphate proportion

A large number of requirements should be satisfied by a potential builder such as: buffer capacity, bleach compatibility and soil deflocculation [40]. The inorganic builder sodium tripolyphosphate (STPP) meets the essential requirements of a builder and therefore it was the most widely used in the past. In addition to its capacity to facilitate dissolution of detergents, STPP can efficiently remove the Ca^{2+} and Mg^{2+} ions presented in hard water and soils, maintains alkalinity during washing, prevents dirt reposing on fabrics by suspending it in the wash-liquor and protects the washing machine against corrosion. Moreover, it shows efficiently performance under all washing conditions [5].

Thanks to these advantages, this builder was used in this study. To determine its effects in removing stain, we varied its proportion in the detergent composition, while fixing a constant percentage of biosurfactant at 10% [5]. We remarked from Table 1 that the rate of oil removal increased with the increase of the amount of STPP to a concentration of 70% beyond which we noted the stability of oil removing (38%). Therefore, a builder

percentage of 70 was needed to obtain the maximum oil removal. This percentage of STPP was selected as an optimum value for further experiments. As remarked, the efficiency of biosurfactant was similar to SDS in removing the oil.

3.1.2 Evaluation of the effect of SPB1 biosurfactant proportion

As surfactants represent one of the most important detergent ingredients, we studied in this part, the effect of its percentage on the oil removal from the pieces of cotton cloth. According to Table 2, the oil removal was negligible when there was no surfactant in the formulation (24%) but it increased rapidly with the increasing of biosurfactant and SDS percentage in the formulation up to 10% to achieve 38 and 39% respectively, indicating the clear effectiveness of biosurfactant in oil removal. This result was in accordance with those reported by Kesting et al., (1996) [27]. Indeed, when they evaluated the detergency activity in removing oils from fibers using *Rhodococcus erythropolis* biosurfactants (trehalosetetraester and trehalosedicorynomycolate), the amount of the oil removed from the fabric was higher compared with the control experiment where a surfactant-free medium was used as washing agent.

However, values greater than 10% of biosurfactant did not improve the oil removal. This behavior may be attributed to aggregation of biosurfactant above its CMC, which made it less effective for scouring [15]. This percentage of surfactant of 10% was selected as the optimum value for further experiments.

3.2 Optimization of washing conditions on removing oil from cotton fabric

The basic prerequisite for any component to be included in laundry detergents should be its stability at the alkaline pH range and the ability to withstand the washing temperature [9]. Thus, stability studies under such conditions are important to determine the viability of possible use of some biosurfactants [41]. The crude lipopeptide biosurfactants from *Bacillus subtilis* used in this study were very stable over a considerable pH range from 2 to 9. Moreover, it retained more than 80% and 50% of its activity after 60 min of incubation at 70 and 80°C, respectively [32]. This pH stability and thermal resistance of *B. subtilis* biosurfactants favor their inclusion in laundry detergent formulations.

Three formulations were tested. The first one was exempt from surfactant (SS). The second and the third formulations contained, respectively, 10% of biosurfactant (BIO) and SDS.

3.2.1 The effect of pH

The pH of the solution is an important chemical property which could act on the solubility of the soiling in the cleaning agent. Figure 1 represents the oil removed from cotton pieces as a function of the pH of washing solution. As observed, biosurfactant and SDS had a maximum effect at a neutral pH of the order of 38 and 39%, respectively. As reported by Benincasa et al., (2010) [42] and Ozdemir et al., (2004) [43], at neutral pH, the

Figure 1. Effect of pH on the removal of oil from cotton cloth (temperature = 25°C; stirrer velocity = 750 RPM; washing time = 20 min) (Each value represents mean $\pm$ standard deviation ($n = 2$)).

molecules of the major ionic surfactants present higher critical micelle concentration. That means more compact layers of the surfactant were formed at the oil water interface resulting in the displacement of more oil molecules from the cloth. Our results corresponded to those described by Bafghi and Fazaelpoor (2012) [5] showing that rhamnolipids used in laundry detergent, under the same conditions, had an optimum performance in removing oil from the cloth (32%) at neutral pH.

3.2.2 The effect of stirrer velocity

Dirt adheres to the textile fibers with a certain energy that must be overcome to get rid of. We must therefore implement a mechanical action to circulate water containing detergent between the fibers of the cloth to allow the stall and training of dirt. The effect of the stirrer velocity on the oil removal from the pieces of cotton cloth was presented in Fig. 2. As expected, oil removal increased with stirrer velocity. At 1000 RPM, the percentage of removing oil achieved 50% when using biosurfactant while it was only 41% when using SDS. Therefore, this result indicated the washing performance of these biomolecules.

Figure 2. Effect of stirrer velocity on the oil removal from cotton cloth (pH = 7; temperature = 25°C; time = 20 min) (Each value represents mean $\pm$ standard deviation ($n = 2$)).

Figure 3. Effect of temperature on the removal of oil from cotton cloth (pH = 7; stirrer velocity = 1000 RPM; washing time = 20 min) (Each value represents mean $\pm$ standard deviation ($n = 2$)).

3.2.3 The effect of temperature

The effect of temperature on the oil removal from the pieces of cotton cloth was presented in Fig. 3. In order to save the energy resulting from heating water used for washing, the ability of detergent components to perform wash function at lower (room) temperature should be addressed [44]. However, Sajna et al., (2013) [22] proved that high washing temperature result in better cleaning and Bafghi and Fazaelpoor (2012) [5] demonstrated that higher temperature causes better dissolution of oil in water. This result was in accordance with those reported here. In fact, we note the effectiveness of temperature in oil removal. It could be seen that oil removal increased with the increase of temperature until it reached 65°C to achieve 58%, while temperature higher than 65°C did not improve the oil removal. Therefore, it was selected as the optimum value for further experiments. Moreover, this figure clearly demonstrates the effectiveness of biosurfactant comparing to the SDS in oil removing at high temperature.

3.2.4 The effect of washing duration

The washing duration can be estimated from the amount and the type of stain and also from the temperature. Generally, a higher wash temperature decreases the cleaning time. In previous study, using sophorolipids biosurfactant produced by *C. bombicola* (ATCC22214) as a stain cleaner could reduce the soaking time required during washing process, thus making the interface for stain removal available within shorter period [28]. Figure 4 shows the effectiveness of SPB1 biosurfactant on oil removal, comparing to SDS when working at 65°C and varying washing duration from 10 min up to 60 min. It was clear that oil removal increases with the increase of washing time. After 60 min, the removal rate reach 62 and 75% when using respectively, SDS and SPB1 biosurfactant showing its effectiveness in oil removal.

All these results confirmed the effectiveness of using the biosurfactant of *B. subtilis* SPB1 as surfactant agent in biological washing powders at a percentage of 10% and this at a neutral pH, a temperature of 65°C, a speed agitation of 1000 RPM and a washing time of 60 min.

Figure 4. Effect of washing time on the oil removal from cotton cloth (pH = 7; temperature = 25°C; stirrer velocity = 1000 RPM) (Each value represents mean $\pm$ standard deviation ($n = 2$)).

3.3 Comparison of cleaning efficiency

3.3.1 Comparison with chemical surfactant

The cleaning efficiency of SPB1 biosurfactant formulation was compared to that prepared using a chemical surfactant, the SDS. This later, being anionic surfactant, acts both as an emulsifier and a detergent. It picks hydrophobic substances from fabric surface, emulsify them and suspend water insoluble impurities in the aqueous media [13]. The Fig. 5 is the representative of detergency test demonstrating comparative washing performances of SPB1 biosurfactant, SDS, commercial detergent, and bio-detergent combination against oil and tea stains. Tap water was taken as control. In fact, as observed, the chemical surfactant formulation exhibited less cleaning efficiency on stains removing comparing to SPB1 biosurfactant formulation. It removed only 62% of oil and 75% of tea while the bio-washing could, respectively, eliminate more than 75 and 80% of stains. These results are in accordance with those obtained by Turbekar et al., (2014) [13] who proved that the cleaning efficiency of rhamnolipid, obtained from *Pseudomonas aeruginosa*, formulation was equivalent to tow conventional synthetic surfactants on removing white board marker stains. Moreover, a thermostable biopolymer produced by an alkaliphilic bacterium,

Figure 5. Comparison of the effect of certain formulations for the stain removal from fabric cotton (pH = 7; temperature = 65°C; stirrer velocity = 1000 RPM; time = 60 min).

Klebsiella sp. strain RJ-03, composed of sugar, uronic acid, protein and sulfate, showed an excellent oil removing efficiency from soil and cotton cloths as compared to chemical surfactants [45]. Hence, biosurfactant could be used to substitute synthetic chemical as they are more biocompatible to the environment.

3.3.2 Comparison with commercial detergent

In the detergency test, stains differ in their chemical nature. They could be notorious such as caffeic acid, yellow solid containing phenolic, acrylic group in coffee stain, and curcuminoids in turmeric. These kinds of tough stains could be removed by using bleach or acids which could damage the fabric. On the other hand, biosurfactant can work as well as detergent since there is an indication that they are able to remove the majority of stains [28]. For this, it will be important to compare the SPB1 biosurfactant formulation efficiency with one commercial detergent available in the Tunisian market. Although the commercial detergent contains more additives, the stain removal by crude biosurfactants alone was more efficient. From Fig. 5, 75% and 81% of removing motor oil and tea, respectively, were found when using biosurfactant while they were only 34% and 58%, respectively, when using commercial detergent.

Many researchers focused on the efficiency of biosurfactant to clean stains. In fact, Sajna et al., (2013) [22] reported that stain removal by glycolipides biosurfactants, produced by *Pseudozma sp* NII 08165, alone was efficient and comparable to that of the commercial detergent. It could remove more than 80% of goot blood, ketch up and chocolate from cotton fabric. Moreover, Bafghi and Fazelipour (2012) [5] described that although the biosurfactant formulation is less effective than the commercial products in stain removal from cotton cloth, the difference is not very high. They noted that bio-washing can remove 30% of oil and more than 85% of chocolate and albumin from cotton clothes while commercial detergent can remove 53, 87, and 92%, respectively. Besides, as investigated by Joshi-Navare et al., (2013) [28], the performance of Sophorolipids (SLs), a glycolipidic biosurfactant produced by *C. bombicola* (ATCC22214) was nearly equal to the detergent in remaining four types of stains (coffee, turmeric, oil, and poster color) from two different types of fabrics, namely, cotton and polyester. However, the removal of oil / blood from cotton fabrics by crude CLP biosurfactants from *B. subtilis* strain was lower than the detergent solution alone [9].

3.3.3 Comparison with combined biosurfactant-commercial detergent

It is well known that certain mixtures of surfactants can provide better performance than pure surfactants for a wide variety of applications [46]. Indeed, Hall et al., (1996) [4] invented a laundry detergent composition containing a combination of sophorolipids and nonionic surfactant. The described detergent compositions had an enhanced oily soil detergency in fabric washing. In this study, one commercial detergent was supplemented with the SPB1 biosurfactant at a ratio of 9:1 (w:w). Studies on CHAL (compost humic acid-like matter) in detergent formulation revealed a proportion of 1:1 (w/w) biosurfactant-commercial surfactants gave significant synergy on wash performance [47].

The results showed that the biosurfactant had a significantly positive effect on the performance of the commercial detergents. We remarked from Fig. 5, that detergent combination resulted in better oil and tea removal, which achieved respectively 45% and 65% against 34% and 58% when using commercial detergent alone. As described by Joshi-Navare et al., (2013) [28], SLJO -detergent combination resulted in better coffee stain removal from cotton fabric. Moreover, Mukherjee (2007) [9] reported that it was evident, from the enhanced removal of oil (9–14%) and blood stain (23–26%), that the washing was performed in the presence of detergents containing CLP biosurfactants, from *B. subtilis*, compared to the detergent solution alone. Therefore, crude biosurfactants can be used as laundry additive as they are environmentally friendly, improve wash performance of the detergent and require less post wash rinsing as they are nontoxic to the skin and reduces energy and water [22].

4 Concluding remarks

It can be concluded that SPB1 lipopeptide biosurfactant can be an effective surfactant in detergent formulations. The performance of SPB1 biosurfactant-containing detergent is comparable with commercial ones especially for the removal of hydrophilic stains. This study has great significance because it is already known that microbial biosurfactants are considered safer alternative to chemical or synthetic surfactants thanks to their lower toxicity and higher biodegradability. Moreover, its additive interaction with the components of tested laundry detergents to improve the wash performance by efficiently removing oil and tea leads us to conclude that the future use of this lipopeptide biosurfactant as laundry detergent additives is highly promising.

Practical application

Highlights

The application of the SPB1 biosurfactant in detergent formulation.

The investigation of washing formulation and condition for strain removing.

A comparative study of washing performance of industrial and BIO-detergent.

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Permission statements

The manuscript does not contain experiments using animals or human studies.

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