Kill•Spill

Integrated Biotechnological Solutions for Combating Marine Oil Spills

Deliverable D5.8

Delivery of surfactants to sediment with releasing formulations

This project is supported by the European Union under the Food, Agriculture and Fisheries and Biotechnology theme of the 7th Framework Programme for Research and Technological Development under GA no. 312139

Work package	WP5 Efficient cleanup of contaminated sediments due to oil spills. Emphasis on biotechnological solution.
Deliverable no	D5.8
Deliverable title	Delivery of surfactants to sediment with releasing formulations.
Due date:	Month 40 (2016-04-30)
Actual submission date:	Month 41 (2016-05-29)
Start date of project:	2013-01-01
Deliverable Lead Beneficiary (Organisation name)	UNIBO
Participant(s) (Partner short names)	UNIBO
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Dissemination Level: (PUblic, Restricted to other Programmes Participants, REstricted to a group specified by the consortium, COnfidential only for members of the consortium)	PU
Deliverable Status:	final

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1 About this deliverable

This deliverable reports on the activities carried out during the months 25-40 in subtask 5.3.2. During the first 24 months of the project, a number of biogenic non-toxic and biodegradable pollutant mobilizing agents and surfactants of plant, animal and microbial origin were investigated in terms of their capability to stimulate the aerobic and anaerobic biodegradation of the *n*-alkane fraction of oil hydrocarbons in contaminated marine sediments (see deliverable D5.3 for details). The main objectives of the study carried out in months 25-40 were i) to assess the effectiveness of the same pollutant mobilizing agents and surfactants in enhancing the hydrocarbons bioavailability in crude oil contaminated marine sediments and ii) to develop approaches for the deployment to the sediment of the surfactants that were more effective in terms of bioavailability and biodegradation enhancement.

In particular, two types of cyclodextrins having different hydrophilicity, namely hydroxypropyl-β-cyclodextrins (HPB-CD) and randomly methylated β-cyclodextrins (RAMEB-CD), two commercial soy lecithin products (Textrol F and Solec C) having different hydrophilic/lipophilic balances (HLB 4 and 7, respectively), bile acids and two microbial surfactants, namely rhamnolipids and sophorolipids were investigated in bioavailability enhancement tests based on the measurement of hydrocarbons pore water concentration via passive sampling with polydimethylsiloxane (PDMS) fibers. For the surfactants encapsulation tests, HPB-CD and sophorolipids were selected. Two different encapsulation strategies were followed: i) encapsulation in biodegradable organic polymers, namely polybutylene succinate (PBS) and polylactic acid (PLA), and ii) encapsulation in hydrogels of agar.

Cyclodextrins were the most effective biogenic agents able to increase the *n*-alkanes bioavailability in sediments both immediately after sediment contamination and after hydrocarbons adsorption to the sediment. Lower bioavailability enhancement was exhibited by the soy lecithin Textrol and sophorolipids. HPB-CD was efficiently encapsulated in hydrogels, whereas sophorolipids in PBS microspheres. HPB-CD was quickly released from hydrogel capsules (approx. 3-4 days, depending on agar concentration), whereas slower release was observed for sophorolipids from PBS microspheres (approx. 40 days). Both encapsulated agents affected hydrocarbons porewater concentrations similarly to free agents in a sand freshly contaminated with crude oil.

2 Introduction

Marine sediments represent the final sink for weathered petroleum hydrocarbons after oil spill events, due to their strong tendency to adsorb on the sediment organic matter, where their biodegradation rate is reduced due to their low bioavailability to microorganisms.

Non-toxic and biodegradable pollutant mobilizing agents of plant and animal origin and of microbial surfactants can be used to enhance hydrocarbons bioavailability and biodegradation. In particular, during the first 24 month of the Kill•Spill project we found that supplementation of biogenic agents to anaerobic microcosms of an actually contaminated sediment from Gela stimulates the indigenous anaerobic microbial activity, as revealed by the onset of reducing conditions and sulphate reduction in microcosms. Furthermore, we observed that extensive biodegradation of *n*-alkanes (more than 80%) is promoted in 40 weeks by cyclodextrins (hydroxyl propyl-β-cyclodextrins and randomly methylated β-cyclodextrins), sophorolipids and, to less extent, soy lecithins (Solec C and Textrol F) (see D5.3). Therefore, the observed enhancement of hydrocarbons biodegradation might be due either to the increased bioavailability of hydrocarbons by surfactants or to a biostimulation of the indigenous microbial community by biosurfactants, that might be used as carbon and energy source or, in the case of soy lecithins, as nitrogen and phosphorous source. One of the main aims of the

activities carried out in the following months was therefore to assess the effect of the tested biosurfactants/mobilizing agents on the hydrocarbons bioavailability. While bulk solid-phase concentration is generally used to assess sediment quality but is not useful for measuring the bioavailability (Burton, 1991), i*n situ* porewater concentration is regarded as an effective tool for estimating the bioaccumulation of sediment-associated contaminants and therefore their bioavailability. However, measurement dissolved hydrocarbons concentrations in porewater is often difficult due to the long and rigorous analytical procedures required to obtain large volumes of porewater to detect a sufficient level of hydrocarbons (Mayer et al., 2000). An alternative monitoring approach is the passive sampling of the interstitial water (or porewater) in contaminated sediments (Thomas et al, 2014, Hong et al, 2015). Several passive sampling techniques have been studied, including semi-permeable membrane devices (SPMDs), polyoxymethylsoloxane (POM) sheets, polyethylene (PE) sheets and polydimethylsiloxane (PDMS) fibers. These samplers are directly placed *in situ* until equilibrium is reached and the porewater concentration is then calculated through partition coefficients. Studies on passive samplers showed that a large amount of time could be required to reach the equilibrium, although for PDMS fibers only few days are sufficient to attain the equilibrium of some reference compounds with the porewater (Lampert et al, 2015). PDMS fibers have been therefore selected for monitoring changes in the porewater concentration of oil hydrocarbons produced by the selected biosurfactants/mobilizing agents.

Surfactant releasing formulations for the surfactants delivery to the sediment should be developed in order to prevent the random dispersion of the surfactants in marine water. Polymers that can be rapidly biodegraded in the marine environment should be used for this purpose, in order to obtain an environmental friendly formulation. The double emulsion method has been widely applied mainly for the controlled delivery of drugs (Tapan et al., 2013). Several polyesters can be used to encapsulate active principles, that are released upon hydrolytic or bio-degradation of the polymer. Therefore, a quite slow release rate can be expected, as compared to hydrogels, where release of the encapsulated molecule occurs via diffusion. The latter approach should also be based on biodegradable polymers in the marine environment; for this reason, the use of agar, a natural polysaccharide, has been selected. A successful encapsulation of biosurfactants in biodegradable polymers involves i) high encapsulation efficiency of such compounds in the tested polymers and ii) ii) known release rate and extent of the surfactants from the formulation. These aspects have been investigated in the encapsulation of 2 selected biosurfactants/mobilizing agents in polyester microspheres and hydrogels.

3 Materials and methods

3.1 Bioavailability tests

3.1.1 Experimental set up

The tests were conducted in triplicate in 120 mL serum bottles containing 80 mL of Gela sediment or sand suspended in marine water (20% dry weight/volume) under sterile conditions. The Gela sediment and sand were contaminated in the laboratory with Dansk Blend crude oil (viscosity at 50 °C: 4.04 cSt; density: 810 g/L) to the final concentration of 5 g/kg immediately before the addition of surfactants and fibers. In order to assess if the enhancement of hydrocarbons anaerobic biodegradation observed in the Gela sediment supplemented with mobilizing agents/biosurfactants (see D5.3) is related to an increase in hydrocarbons bioavailability (porewater concentration), the slurries established with Gela sediment were amended with the same surfactants previously studied in the hydrocarbons biodegradation tests, namely: randomly methylated β-cyclodextrins (RAMEB-CD), hydroxylpropyl-β-cyclodextrins (HPB-CD), rhamnolipids, sophorolipids, the commercial soy

lecithin products Solec C and Textrol F and bile acids. In order to study the effect of encapsulation on surfactant release and hydrocarbons porewater concentration, slurries prepared with contaminated sand were used, and only two surfactants that previously were shown to enhance the anaerobic biodegradation of *n*-alkanes to a higher extent were selected (i.e., HPB-CD and sophorolipids). All surfactants, either free or encapsulated, were added at the same final concentration used previously in biodegradation enhancement experiments (0.2 g/L for rhamnolipids and sophorolipids, 1 g/L for all other surfactants; D5.3). Not amended controls were also set up. (Table 1).

Table 1 Experimental set up.

Effect of surfactants on hydrocarbons porewater concentration (Gela sediment slurries)	Effect of encapsulation on surfactants' ability to increase porewater concentration (Sand slurries)
Un-amended control	Un-amended control
Rhamnolipids	Sophorolipids
Sophorolipids	Sophorolipids encapsulated in PBS
Rameb-CD	HPB-CD
HPB-CD	HPB-CD encapsulated in hydrogel
Solec F	
Textrol C	
Bile acids	

3.1.2 Analysis of hydrocarbons bioavailability

The evaluation of hydrocarbons bioavailability was monitored via the measurement of *n*-alkanes concentration in the porewater through passive sampling with polydimethylsiloxane fibers (PDMS, Figure 1). The outer diameter of the PDMS was 558.8 µm and the inner diameter 486 µm (i.e., the thickness annulus of PDMS was 35.4 µm and the fiber volume 0.597 µL/cm).

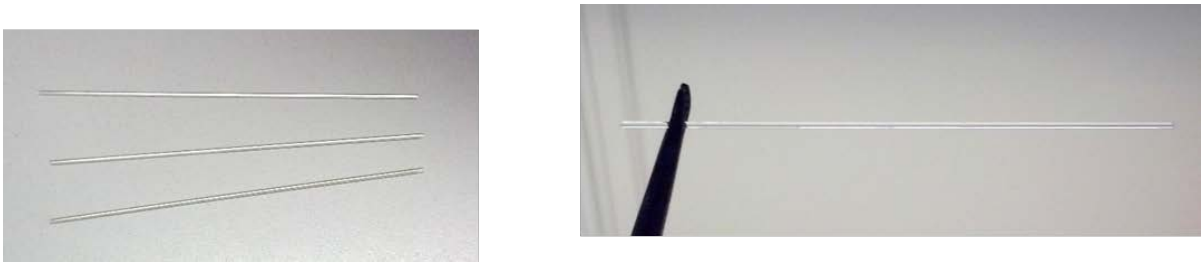

Figure 1 Polydimethylsiloxane fibers.

PDMS fibers (5 cm of length) were incubated in the microcosms at 20 °C, 150 rpm, and replaced every 20 days to sample water-dissolved hydrocarbons. At each sampling, fibers were removed from the microcosms and cut into 1 cm segments; after elution into hexane (100 µL) for 16 h, *n*-alkanes were analysed with an Agilent Technologies gas-chromatograph 6890N equipped with flame ionization detector. The measured concentration of *n*-alkanes in the solvent was converted to total mass and divided by PDMS volume in the 5 cm fibers, in order to determine the *n*-alkanes concentration on fibers (C_{PDMS}). The porewater concentration (C_w) was then obtained by dividing the

concentration on fibers by the fiber-water partition coefficient (K_{PDMS-W}), which is correlated with the octanol–water partition coefficient as follows (Thomas et al., 2014)

$$\log K_{PDMS-W} = 0.725 \log K_{OW} + 0.479 \ (R^2 = 0.99)$$

where K_{OW} is given by SPARC estimates (Table 2).

Table 2 $\log K_{OW}$ and $\log k_{PDMS-W}$ of the different *n*-alkanes (C_{10}-C_{36}).

n-alkanes	$\log K_{OW}$	$\log K_{PDMS-W}$
C10	5.25	4.285
C11	5.74	4.641
C12	6.23	4.996
C13	6.73	5.358
C14	7.22	5.714
C15	7.71	6.069
C16	8.20	6.424
C17	8.69	6.779
Pristane	9.38	7.280
C18	9.18	7.135
Phitane	9.87	7.635
C19	9.67	7.490
C20	10.16	7.845
C21	10.65	8.200
C22	11.15	8.563
C23	11.64	8.918
C24	12.13	9.273
C25	12.62	9.629
C26	13.11	9.984
C27	13.60	10.339
C28	14.09	10.694
C29	14.58	11.050
C30	15.07	11.405
C31	15.56	11.760
C32	16.05	12.115
C33	16.54	12.471
C34	17.03	12.826
C35	17.52	13.181
C36	18.01	13.536

3.2 Encapsulation of surfactants

Sophorolipids and HPB-CD were selected for encapsulation tests in polyesters with the double emulsion method and in agar hydrogels.

3.2.1 Encapsulation in polyesters with the double emulsion method

Polybutylene succinate (PBS), polybutylene succinate co-adipate (PBSA), polylactic acid (PLA) and polycaprolactone (PCL) were chosen as degradable polyesters to encapsulate the surfactants using the double emulsion method (Figure 2). The aqueous biosurfactant solution (1 mL) was emulsified with the polymer organic solution containing glycerol as water soluble plasticizer (Figure 2), at high mixing rates (13000 rpm). The emulsion was then dropped in a water solution of a polyvinyl alcohol (0.1 weight/volume) (S3), an anticoagulant stabilizer, under vigorous mixing (6000 rpm). The obtained microspheres were then washed with water, recovered by filtration and dried at room temperature. 10 mL of chloroform containing 0.2 mL of glycerol were used to dissolve 2 g of polymer (solution S1). Amounts of glycerol were varied in the range 0.2-0.6 mL for the preparation of PLA microspheres. One ml of a 30 g/L solution of sophorolipids and a 200 g/L solution of HPB-CD were used as solution S2.

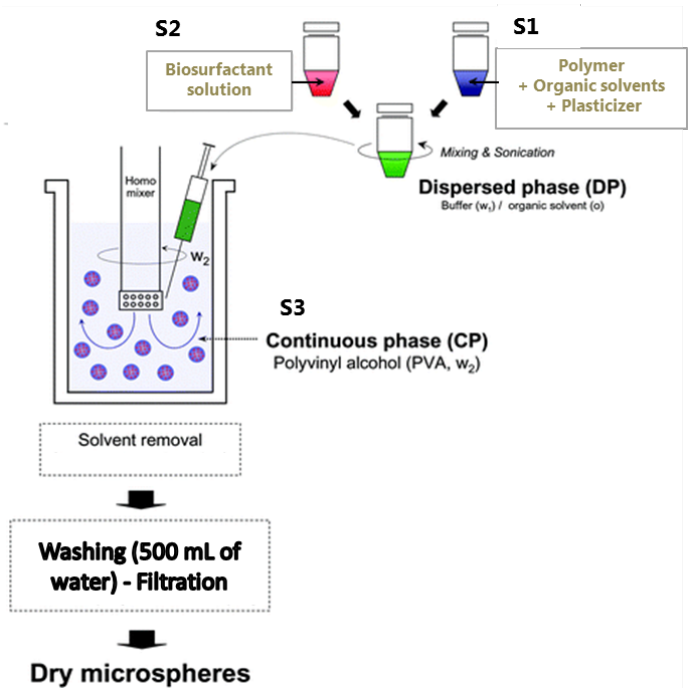

Figure 2 Steps for the encapsulation of surfactants in polyester microspheres using the double emulsion method.

In order to assess the encapsulation efficiency, a known amount of microspheres (0.1 or 0.2 mg) was disrupted and the amount of surfactant released measured chromatographically. Two different approaches were used for the disruption of microspheres containing the two surfactants. The sophorolipids microspheres were suspended in water and subjected to an acid hydrolysis (1.6 M HCl, 95°C, 4h) of both the polymer and sophorolipids; the concentration of the microbial surfactant was then measured after neutralization of the solution with KOH through HPLC-RID analysis (Agilent Technologies 1260 Infinity) of the glucose released. The HPB-CD microspheres were dissolved into chloroform and the released HPB-CD were batch extracted with water (extraction yield 98%). The aqueous phase was recovered and analyzed in HPLC-RID to determine the concentration of HPB-CD.

1 g of PBS microspheres loaded with sophorolipids (28.6 ± 0.8 mg) were incubated statically at 20 °C in 100 mL of sterile water to monitor the surfactants release. The water was periodically sampled

and the sophorolipids release was valued after acid hydrolysis of the water through HPLC analysis of the glucose.

3.2.1 Encapsulation in ager hydrogels

Surfactant water solutions (30 and 50 g/L for sophorolipids and HPB-CD, respectively) were used to dissolve agar at different final concentrations (5, 15 and 50 g/L) by heating at 121°C, 1 atm in autoclave, then poured (200 µL aliquots) in 96 well plates and cooled to room temperature to obtain agar hydrogel capsules loaded with surfactants. Stability of the surfactants to the thermal treatment was verified with agar-free solutions by analyzing the surfactant concentration by HPLC-RID before and after the treatment. In case of sophorolipid analysis, which is based on glucose release from the molecule by acid hydrolysis, the analysis of free glucose concentration in solution was measured before and after the acid hydrolytic step.

96 capsules loaded with HPB-CD were incubated statically at 20 °C in 0.2 L of sterile demineralized and marine water to monitor HPB-CD release through HPLC analysis of the cyclodextrins in the water sampled at different incubation times.

4 Results and discussion

4.1 Effect of free surfactants/mobilizing agents on porewater concentration of *n*-alkanes in Gela sediment

The effect of free surfactants/mobilizing agents on the *n*-alkanes porewater concentration was investigated in Gela sediment spiked with Dansk Blend crude oil (5 g/kg sediment). Spiking was required since no C_{10}-C_{36} *n*-alkanes were detectable anymore in the sediment after prolonged storage (1.5 years approximately, at 4°C), possibly due to the occurrence of extensive degradation by indigenous microbial community. High porewater concentrations of C_{10}-C_{36} *n*-alkanes (ranging from 122 ± 30 to 1183 ± 428 ng/L) were detected in all the slurries after 20 days of incubation, which suggests a weak initial adsorption of *n*-alkanes to the sediment particles, probably due to the sediment spiking immediately before the start of incubation in the presence of fibers (Figure 3). However, a remarkable decrease of C_{10}-C_{36} *n*-alkanes porewater concentration was detected in the un-amended controls after 40 days (5.3 ± 0.0 ng/L) and 60 days (8.7 ± 7.8 ng/L) of incubation. This indicates that adsorption of *n*-alkanes to the sediment particles was completed and that equilibrium was reached after 20 days of incubation. Porewater concentrations similar to those detected in the un-amended controls were measured after 40 day and 60 days in slurries amended with sophorolipids, rhamnolipids, Solec and bile acids, thus indicating that these surfactants does not affect significantly the adsorption of *n*-alkanes to the sediment and thus their bioavalability. Conversely, porewater concentrations of *n*-alkanes remarkably higher that those detected in the un-amended controls were observed in the slurries amended with both cyclodextrins and Textrol after 40 days and 60 days of incubation, being porewater concentrations higher at day 40 than day 60. This indicates that adsorption of *n*-alkanes to the sediment was still occurring, and therefore that cyclodextrins and Textrol are able to reduce *n*-alkane adsorption rate, and thus to increase the porewater concentration and bioavailability of freshly spiked *n*-alkanes (Figure 3). This also suggest that the application of these biosurfactants/mobilizing agents might be suitable right after oil spill occurrence.

To test their effect on adsorbed *n*-alkanes, after day 60, i.e. after adsorption of spiked *n*-alkanes was completed and equilibrium reached, RAMEB-CD, HPB-CD and Textrol F were added to the un-amended controls, sophorolipids and rhamnolipids microcosms, respectively. Only cyclodextrins were able to enhance *n*-alkanes porewater concentration (Figure 4). This indicates that cyclodextrins

capability to stimulate the *n*-alkanes anaerobic biodegradation observed in the previous studies (D5.3) was probably due to their capability di increase *n*-alkane bioavailability. Conversely, sophorolipids, as well as soy lechihins Solec and Textrol, which also were able to stimulate the anaerobic hydrocarbons biodegradation, could not increase significantly the bioavailability of adsorbed *n*-alkanes and likely acted mainly as biostimulants used as carbon and energy or nitrogen and phosphorous sources by the sediment indigenous microorganisms.

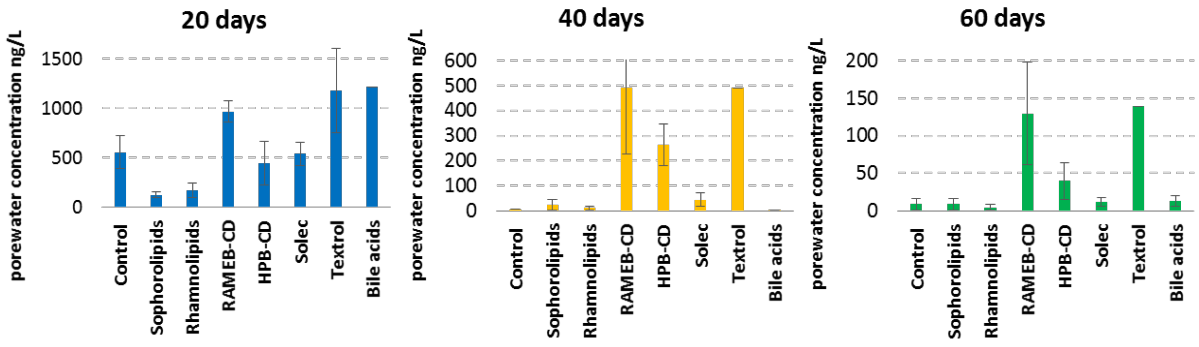

Figure 3 **Porewater concentration (ng/L) of C_{10}-C_{36} *n*-alkanes in sterile Gela sediment slurries spiked with Dansk Blend crude oil in presence of different surfactants after 20, 40 and 60 days of incubation.** Data are the mean of triplicate incubations (± standard deviation).

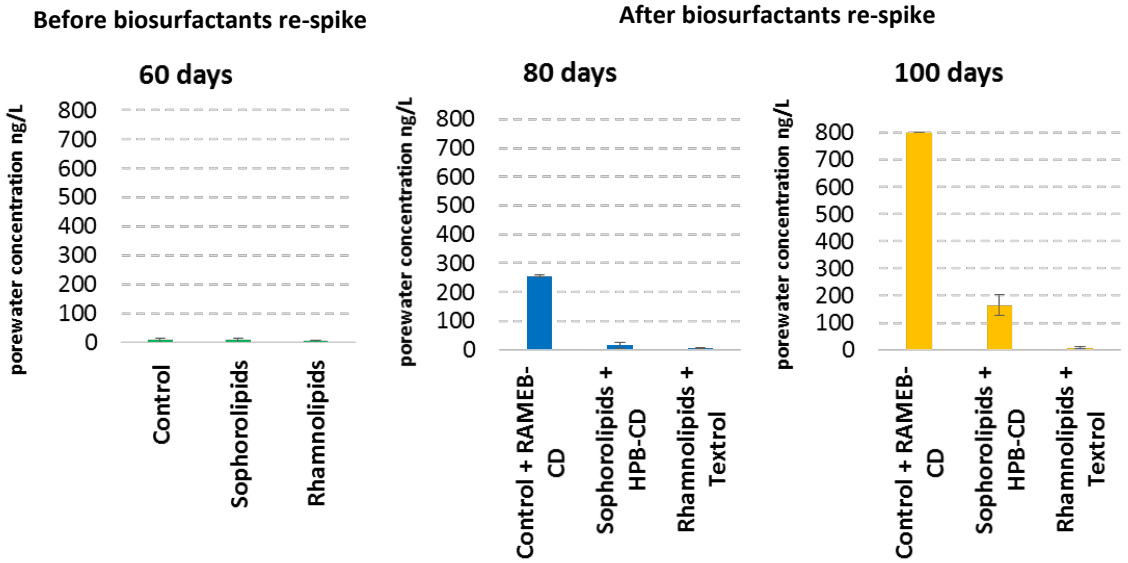

Figure 4 **Porewater concentration (ng/L) of C_{10}-C_{36} *n*-alkanes in un-amended controls, sophorolipids and rhamnolipids Gela sediment slurries before (60 days of incubation) and after (80 and 100 days of incubation) the addition of RAMEB-CD, HPB-CD and Textrol F, respectively.** Data are mean of triplicate incubations (± standard deviation).

4.2 Surfactant encapsulation

Cyclodextrins were selected for encapsulation tests given their capability to enhance both *n*-alkanes bioavailability and biodegradation. In particular, since a suitable HPLC-RID method could be set up only for the quantitative analysis of HPB-CD at concentrations down to 1 mg/L, only HPB-CD were used. In addition, the encapsulation of sophorolipids was also investigated, since their marked

capability to stimulate the anaerobic biodegradation (approx. 80% of *n*-alkanes removal, D5.3) makes this microbial surfactant a potential candidate as bioremediation agent for marine sediments contaminated by oil spills.

4.2.1 Surfactants encapsulation in polyesters

Sophorolipids could be efficiently encapsulated in PBS microspheres. From each encapsulation batch, approximately 1-1.4 g of PBS microspheres containing 28.6 ± 0.8 mg of sophorolipids/g of spheres could be obtained (Figure 5), corresponding to an encapsulation efficiency of 95% (Table 3).

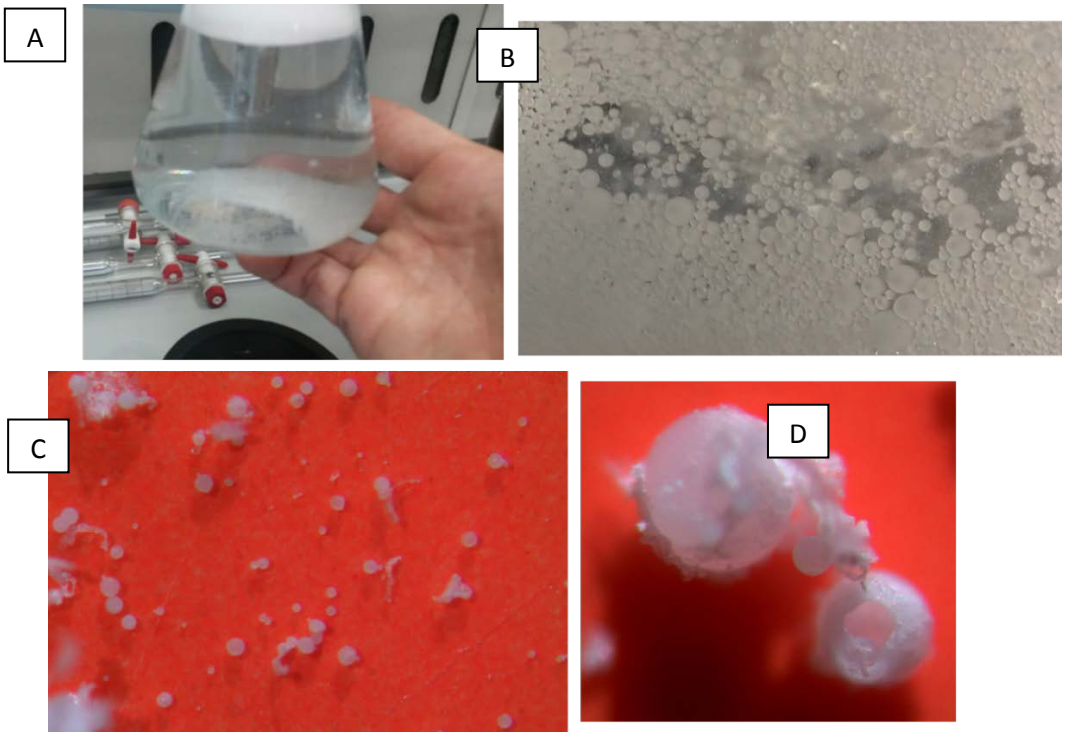

Figure 5 **Microspheres of PBS loaded with sophorolipids: washing step (A-B) and stereo microscope image of dried microspheres (C-D). The empty cavity of one broken microsphere is visible in panel D.**

Table 3 **Loss of surfactants during preparation of microspheres and efficiency of encapsulation in polymers. HPB-PLA_A, HPB-PLA_B and HPB-PLA_C are samples prepared with 0.2, 0.4, 0.8 g of glycerol in solution S3, respectively.**

Sample (surfactant, polymer)	Amount of HPB-CD detected in the water phase before washing (%)	Amount of HPB-CD detected in the water phase after washing (%)	Amount of HPB-CD recovered from microspheres (%)
Sophorolipids, PBS	0	0	95
HPB-CD, PBS	30	90	7
HPB-CD, PLA_A	36	92	2
HPB-CD, PLA_B	27	93	0
HPB-CD, PLA_C	60	100	0

Conversely, the encapsulation efficiency of HPB-CD in PBS and in PLA was remarkably lower, and most the HPB-CD initially used for sphere preparation was detected in the water solution either before washing, as a consequence of low encapsulation, or after washing, probably due to microsphere breakage (Table 3). Attempts to increase the breaking strength of PLA microspheres by increasing the amount of plasticizer (glycerol) in the polymer solution up to 0.8 g/L were unsuccessful (Table 3).

Therefore, only sophorolipids could be efficiently encapsulated in polyester microspheres, namely PBS. Monitoring of sophorolipids during static incubation of 1 g of sophorolipids-PBS microcapsules in 100 mL water at 20 °C revealed that 86% of the surfactant is released from microspheres after 38 days of incubation (Figure 6).

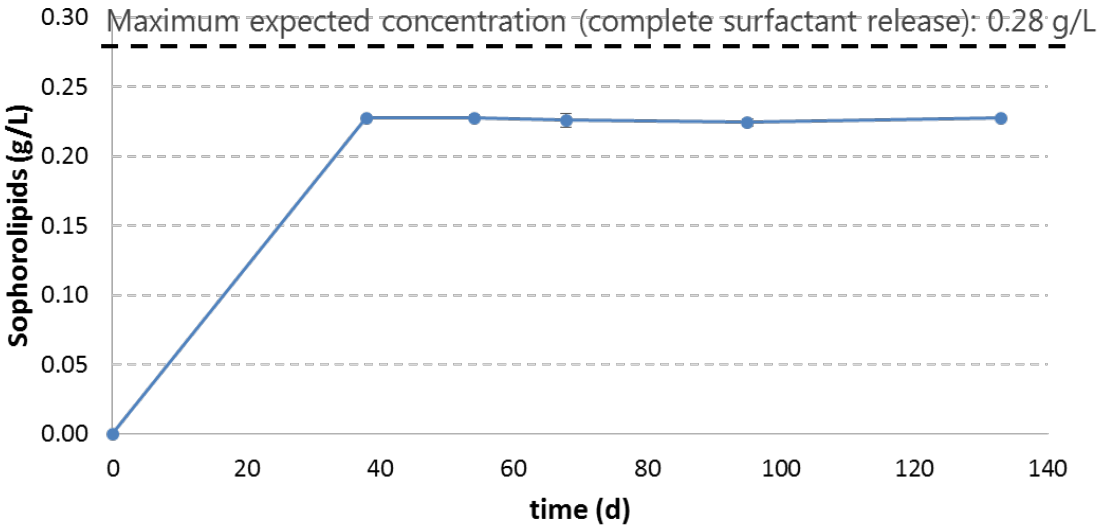

Figure 6 Sophorolipids (g/L) release from PBS microspheres in sterile marine water over time (days).

4.2.1 Surfactants encapsulation in hydrogels (agar)

Sophorolipids (30 g/L) prevented the agar polymerization at all agar concentrations tested (5, 15, 50 g/L), while HPB-CD did not (Figure 7). Thus, the surfactant release from hydrogel capsules (96) in 100 mL of marine and demineralized water under static conditions at 20°C was monitored only for HPB-CD.

Figure 7 Capsules of agar (50 g/L) loaded with HPB-CD (200 g/L).

The concentration of agar in the capsules influenced HPB-CD release in marine water; increasing agar concentration reduced the release of HPB-CD. Depending on agar concentration, approximately 50-70% of HPB-CD was released within the first 8 hours of incubation at 20 °C. In demineralized water the release of cyclodextrins from the hydrogel capsules was more rapid and less influenced by the agar concentration (Figure 8).

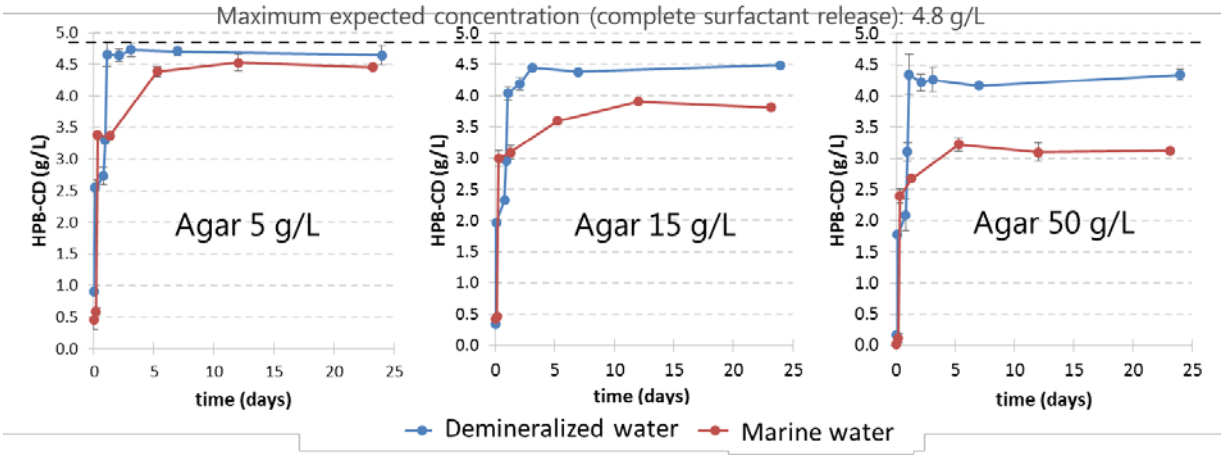

Figure 8 **Concentration HPB-CD (g/L) released over time in sterile marine and demineralized water from capsules with different concentration of agar (5, 15 and 50 g/L).**

4.3 Effect of encapsulation on surfactants' ability to increase porewater concentration of *n*-alkanes in sand slurries

The effect of encapsulation of the ability of HPB-CD to increase the *n*-alkanes porewater concentration was investigated in a sand spiked with Dansk Blend crude oil (5 g/kg) suspended at 20% in seawater under sterile conditions supplemented with free and encapsulated agent. Although sophorolipids were not able to increase *n*-alkane porewater concentration in sediment slurry, free sophorolipids and sophorolipids encapsulated in PBS microsphere were also tested, in order to investigate their potential capability to increase *n*-alkane porewater concentration in a different matrix that is expected to adsorb more weakly *n*-alkanes. High and similar porewater concentrations of C_{10}-C_{36} *n*-alkanes (ranging from 322 ± 84 to 467 ± 88 ng/L) were detected in all the slurries after 20 days of incubation, as detected previously in the spiked Gela sediment (Figure 9), regardless of the addition of free or encapsulated surfactants. After 40 days of incubation, a remarkable decrease of *n*-alkanes porewater concentration (125 ± 11 ng/L) was observed in the un-amended controls, confirming that adsorption was taking place, although at lower rate than in Gela sediment. Conversely, the hydrocarbons porewater concentration increased in the microcosms amended with free HPB-CD (2293 ± 231 ng/L) and sophorolipids (956 ± 336 ng/L), indicating that desorption started to take place earlier than in Gela sediment and also in the presence of sophorolipids (Figure 9). This indicates that also sophorolipids might increase *n*-alkane bioavailability in matrixes containing lower amounts of organic matter. Remarkably, *n*-alkanes porewater concentrations similar to those measured with free HPB-CD were detected in the presence of hydrogel-encapsulated HPB-CD (2557 ± 141 ng/L). *n*-alkane porewater concentrations higher than in the un-amended controls were detected also in the presence of PBS-encapsulated sophorolipids (465 ± 190 ng/L), although these were approximately 50% of those measured in the presence of free sophorolipids (Figure 9). Indeed, when compared to the addition of the corresponding free agents, the encapsulation of sophorolipids in PBS microspheres (sophorolipids) resulted in lower porewater concentrations than

encapsulation of HPB-CD in agar hydrogel; this is consistent with the lower release rate of sophorolipids from PBS microsphere than of HPB-CD from agar hydrogel capsules. Overall, these data indicate that the both formulations might be effective solutions for the delivery and release of surfactant/mobilizing agents able to increase hydrocarbons bioavailability and biodegradation in marine sediments.

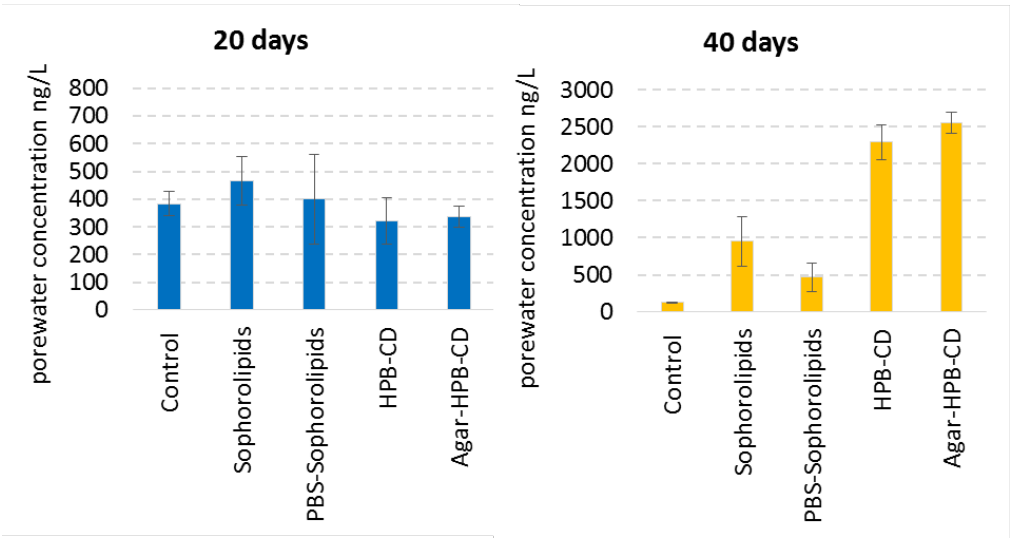

Figure 9 **Porewater concentration (ng/L) of C_{10}-C_{36} *n*-alkanes in un-amended controls and in slurries supplemented with the free and encapsulated surfactants (sophorolipids and HPB-CD) after 20 and 40 days of incubation. Data are mean of triplicate incubations (± standard deviation).**

5 Conclusion

Two cyclodextrins, RAMEB-CD and HPB-CD, are the most effective agents, among those tested, able to increase porewater concentrations of *n*-alkane hydrocarbons, both immediately after contamination as well as after complete adsorption of hydrocarbons to sediment has occurred. A lower effect on hydrocarbons bioavailability is exerted by the commercial soy lecithin Textrol, which only reduces hydrocarbons adsorption to sediment, and by sophorolipids, that are effective only on sand matrixes exhibiting a lower tendency to adsorb hydrocarbons. Since all these agents were previously found to stimulate the anaerobic biodegradation of *n*-alkanes in Gela sediments, their hydrocarbons biodegradation enhancement activity is likely associated to different phenomena, i.e., increase of bioavailability for cyclodextrins and biostimulation for Textrol and sophorolipids.

HPB-CD could be efficiently encapsulated in agar hydrogel capsules, whereas sophorolipids in PBS microspheres. The release rate from agar hydrogel capsules is higher (50-70% release within the first 8 hours) than from PBS microspheres (86% release within 40 days) and can be partially modulated by changing the agar concentration. Both encapsulation methods allow delivering surfactants to sediments with effects on hydrocarbons porewater concentration comparable or slightly lower than those of the free agents, depending on the agent release rate from the formulation. Therefore, both formulations might be effective solutions for the delivery and release of surfactant/mobilizing agents able to increase hydrocarbons bioavailability and biodegradation in marine sediments.

6 References

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