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Effect of a commercial alcohol ethoxylate surfactant ($C_{11-15}E_7$) on biodegradation of phenanthrene in a saline water medium by *Neptunomonas naphthovorans*

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Abstract

Biodegradation of poorly soluble polycyclic aromatic hydrocarbons (PAHs) has been a challenge in bioremediation. In recent years, surfactant-enhanced bioremediation of PAH contaminants has attracted great attention in research. In this study, biodegradation of phenanthrene as a model PAHs solubilized in saline micellar solutions of a biodegradable commercial alcohol ethoxylate nonionic surfactant was investigated. The critical micelle concentration (CMC) of the surfactant and its solubilization capacity for phenanthrene were examined in an artificial saline water medium, and a type of marine bacteria, *Neptunomonas naphthovorans*, was studied for the biodegradation of phenanthrene solubilized in the surfactant micellar solutions of the saline medium. It is found that the solubility of phenanthrene in the surfactant micellar solutions increased linearly with the surfactant concentrations, but, at a fixed phenanthrene concentration, the biodegradability of phenanthrene in the micellar solutions decreased with the increase of the surfactant concentrations. This was attributed to the reduced bioavailability of phenanthrene, due to its increased solubilization extent in the micellar phase and possibly lowered mass transfer rate from the micellar phase into the aqueous phase or into the bacterial cells. In addition, an inhibitory effect of the surfactant on the bacterial growth at high surfactant concentrations may also play a role. It is concluded that the surfactant largely enhanced the solubilization of phenanthrene in the saline water medium, but excess existence of the surfactant in the medium should be minimized or avoided for the biodegradation of phenanthrene by *Neptunomonas naphthovorans*.

Introduction

Bioremediation has attracted great interest in recent years in decontamination of soil and water polluted by various organic compounds or wastes. However, the success of bioremediation is largely limited in the case of poorly soluble polycyclic aromatic hydrocarbons (PAHs). These substances, originated predominantly from processes such as combustion, are of major environmental or public health concern as they are either known or suspected carcinogens or mutagens (Mizesko et al. 2001; Tsai et al. 2001). Due to their strong sorption to soil particles and low water solubility, PAHs, especially the ones with high molecular weight, are not normally biologically available for biodegradation even though many microorganisms may be able to degrade them. Stimulated by the high solubilization capacity of surfactants for hydrophobic compounds, a new technology, surfactant-enhanced bioremediation, has become a research focus for PAH biodegradation (Mulligan et al. 2001).

Although the conclusions have been unanimous concerning the enhancement of the apparent solubility of hydrocarbons in surfactant micellar solutions from contaminated soil (Grimberg & Aitken 1995; Johnson et al. 1999), conflicting results have been reported on the ability and extent of surfactants in enhancing the biodegradation process. One of the arguments is whether the solubilization of hydrocarbons in micellar solution is conducive or inhibitory to microbial uptake of the hydrocarbons. The inhibitory effect was observed at surfactant concentrations approaching or exceeding the critical micelle concentrations (CMC) and was attributed to factors such as surfactant being toxic to microorganisms (Tsomides et al. 1995), surfactant being used as preferable substrates (Laha & Luthy 1992), and surfactant preventing direct contact between cells and hydrocarbon surface (Efroymson & Alexander 1991). It was also reported that the effect of a surfactant can be bacteria-specific (Allen et al. 1999), which means that the specific interaction between bacteria and surfactant also plays an important role in biodegradation of hydrocarbons. In addition, the bioavailability of PAHs solubilized in surfactant micellar solutions has been reported to vary with the types of surfactants and to be a function of the surfactant concentrations (Guha & Jaffé 1996).

With the increase of environmental awareness, stricter regulations have also been posed for the use of environmentally acceptable surfactants. In this work, a commercially available biodegradable nonionic surfactant (trade name: Tergitol 15-S-7) was used in the study of surfactant-enhanced biodegradation of a model PAH, phenanthrene. The surfactant is a mixture of linear secondary alcohol ethoxylates (chemically represented as $C_{11–15}E_7$, where $C_{11–15}$ indicates the number of carbons in the alkyl hydrophobic part of the molecule, and E_7 the average number of ethoxylate groups in the hydrophilic part), and it is more environmentally benign and more readily biodegradable than the traditionally used alkyl-phenol-polyethoxylate surfactants that have been shown to resist biodegradation and leave relatively stable metabolites that are toxic to both marine and fresh water microorganisms (Van der Meeren & Verstraete 1996). Selection of the linear alcohol ethoxylates is also based on their known high solubilization capacity (Li & Chen 2002). Although many surfactants have been studied for their effects on the biodegradation of PAHs (Grimberg et al. 1996), the use of Tergitol 15-S-7 surfactant in PAH biodegradation, especially with a new type of marine bacteria – *Neptunomonas naphthovorans*, in a saline water environment has seldom been reported.

Materials and methods

Materials

Phenanthrene was purchased from Aldrich Chemicals Co. The commercial nonionic biodegradable surfactant Tergitol 15-S-7 was supplied by Union Carbide (USA). The bacteria, *Neptunomonas naphthovorans* (ATCC 700638), used in the biodegradation experiments, were obtained from American Type Culture Collection (ATCC), and they can aerobically degrade polycyclic aromatic hydrocarbons (Hedlund et al. 1999). An artificial saline water medium, based on the ionic compositions of seawater (Dyksterhouse et al. 1995), was used in the biodegradation experiments. The pH of the medium was adjusted with 2 N hydrochloric acid to 7.5 ± 0.1.

Analytical and experimental methods

Phenanthrene concentration. The separation and quantification of phenanthrene in the surfactant micellar solutions was conducted with a Shimadzu HPLC system equipped with an RF-10AXL fluorescence detector, following the analytical method reported by Li & Chen (2002). Triplicate analyses for each sample were conducted and the average value from the three measurements was used in the paper. The reproducibility of the analyses was quite high, with errors in the range of less than 2%. The presence of the surfactant did not interfere with the detection of phenanthrene because the surfactant cannot render any fluorometric signals.

Biomass. A Shimadzu TOC-5000A total organic carbon analyzer was used for the determination of biomass. The oven temperature was set at 680 °C. The biomass in a solution sample was expressed by the difference of the TOC values measured from the sample before and after centrifugal treatment with an Eppendrof Model

5870R centrifuge operating at 10,000 rpm for 10 min.

CMC determination. The critical micelle concentration (CMC) of the surfactant was determined with a Surface Tensiometer (Krüss drop shape analysis system DSA 10). To determine the CMC of the surfactant in the saline water medium, concentrated surfactant solutions and a series of dilutions were prepared. The surface tensions were measured with the Surface Tensiometer. The CMC value for the surfactant was estimated by plotting the surface tension data against the logarithms of the surfactant concentrations. The surfactant concentration at the transition between the descending line for surfactant concentrations less than CMC and the other line for surfactant concentrations greater than CMC was taken as the CMC value. In the following discussion, the medium with the surfactant concentration above the CMC will specifically be referred to as micellar solution, but, in many cases, the medium and the micellar solution will be used interchangeably.

Phenanthrene solubilization equilibrium. The solubilization experiments of phenanthrene in the surfactant micellar solutions were carried out in 500 ml conical glass flasks, in the method similar to that used by Grimberg et al. (1995). Each of these solubilization flasks was welded with a cylindrical glass well (3.8 cm in id; 1 cm in height) at the center of its bottom. About 10 g of phenanthrene solids was first placed in the glass well and then 200 ml of the micellar solution with a known surfactant concentration was added into the flask. The solubilization flasks were subsequently placed in a thermostatic water bath shaker operated at 150 rpm and 22 °C for 48 h to allow the solubilization of phenanthrene in the micellar solutions to reach equilibrium. The glass well in each of the solubilization flasks functioned to retain phenanthrene during the solubilization experiments, and also to allow different phenanthrene concentrations in the medium to be obtained by controlling the dissolution time when necessary.

Biodegradation of surfactant. The mineral solutions with different surfactant concentrations were prepared by dissolving different amounts of the surfactant in 500 ml conical glass flasks, each of which had 200 ml of the saline water medium mentioned earlier. Bacteria cultured with Tergitol 15-S-7 were harvested at the late exponential growth stage through a centrifugal operation. The bacteria were then re-suspended in a certain amount of the mineral solution and aliquots of the suspension were inoculated into each of the flasks with the prepared surfactant mineral solutions. The flasks were placed in a water bath shaker operated at 150 rpm and 22 °C for biodegradation of the surfactant to take place. Un-inoculated surfactant mineral solutions were also used in the study as abiotic control tests. At various time intervals, samples were taken from each of the flasks, centrifuged at 10,000 rpm and at room temperature (22 °C) for 10 min to remove the bacteria, and then quantified with a TOC analyser for surfactant concentrations.

Biodegradation of phenanthrene. The biodegradation experiments were also carried out in 500 ml conical glass flasks with 200 ml of the prepared saline medium in each. To study the effect of micellar solubilization of phenanthrene on its biodegradation, solutions with the same initial phenanthrene concentration but different surfactant concentrations were inoculated with the same mass of bacteria cultures harvested at the late exponential growth stage. The contents in the flasks were stirred in a thermostatic water bath shaker operated at 150 rpm and 22 °C, and samples were taken from the flasks at various time intervals for the analyses of phenanthrene concentrations. For each solution, an abiotic control test (with the sterilized un-inoculated solution) was also carried out, and it was found that there was no significant abiotic loss of phenanthrene.

Similar experiments were conducted for biodegradation of phenanthrene in the micellar solutions with different initial phenanthrene concentrations but the same surfactant concentration. Different initial phenanthrene concentrations were obtained by controlling the dissolution time of phenanthrene in the solubilization flasks. Two duplicated tests were carried out for all conditions studied and the average from the two duplicates was reported in this paper.

Results and discussion

CMC and solubilization capacity

The measured CMC of the surfactant and the solubility of phenanthrene, C_{CMC}, in the saline

medium without the surfactant or at CMC of the surfactant are given in Table 1. The solubilization capacities of the surfactant for phenanthrene in the medium at surfactant concentrations above its CMC are shown in Figure 1. It can be found that the solubility of phenanthrene in the medium without the addition of the surfactant was 0.70 mg/l, and the solubility at CMC of the surfactant was 1.15 mg/l. Hence, the enhancement of the solubility of phenanthrene in the medium by adding the surfactant at concentrations below its CMC was relatively low (0.013 mg phenanthrene/mg surfactant). When the surfactant concentrations in the medium increased to a value above the CMC, the solubility of phenanthrene in the medium increased linearly with the addition of the surfactant in the medium. The mass or weight solubilization ratio (WSR), which equals to the slope of the curve in Figure 1, became 0.038 mg phenanthrene/mg surfactant.

Table 1. CMC of the surfactant and solubilization data for phenanthrene in the medium

CMC (mg/l)	C_W (mg/l)	C_{CMC} (mg/l)	WSR (mg/l/mg/l)	K_{mw} (l/mg)
32	0.70	1.15	0.038	0.033

CMC – critical micelle concentration of the surfactant; C_W – Solubilization capacity of phenanthrene in the medium in the absence of the surfactant; C_{CMC} – solubilization capacity of phenanthrene in the mineral medium at CMC of the surfactant; WSR – mass (or weight) solubilization ratio, defined as the amount of phenanthrene solubilized by unit weight of surfactant above its CMC in the medium; K_{mw} – micelle–water partition coefficient.

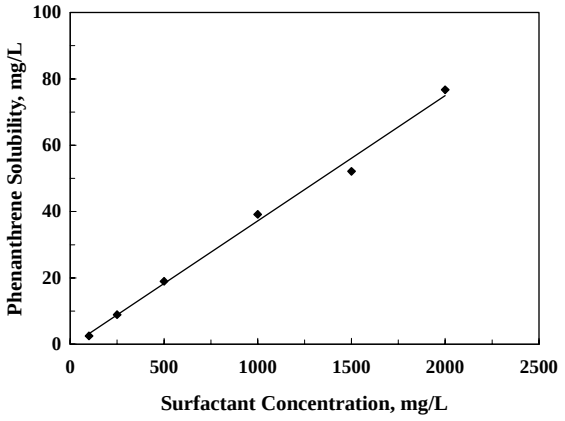

Figure 1. Solubilization capacity of surfactant Tergitol 15-S-7 for phenanthrene in the micellar solution at surfactant concentrations above CMC.

Biodegradation of surfactant

The results of biodegradation of the surfactant at concentrations above the CMC (32 mg/l) are shown in Figure 2. The results indicate that the surfactant was indeed biodegradable. The biodegradability of the surfactant was observed to increase when the surfactant concentration was increased from *ca.* 100 to 200 mg/l, but decreased with further increase of the surfactant concentrations. The mechanism of the inhibition effect at high surfactant concentrations may be due to the change of the micellar structures at higher surfactant concentrations. With the increase of surfactant concentrations, more structured and packed micelles may be formed, which could hinder the close contact between the micellized surfactant and the bacteria (Doong & Lei 2003). The reduced biodegradability of the surfactant at high concentrations was also in agreement with the observed bacterial growth. As shown in Figure 3, the bacterial growth was indeed inhibited when the surfactant concentrations were further increased from 200 to 400 and 600 mg/l.

Effect of surfactant concentrations on biodegradation of phenanthrene

Figure 4 shows the results of biodegradation of phenanthrene in the medium without the surfactant or with the surfactant at a concentration of

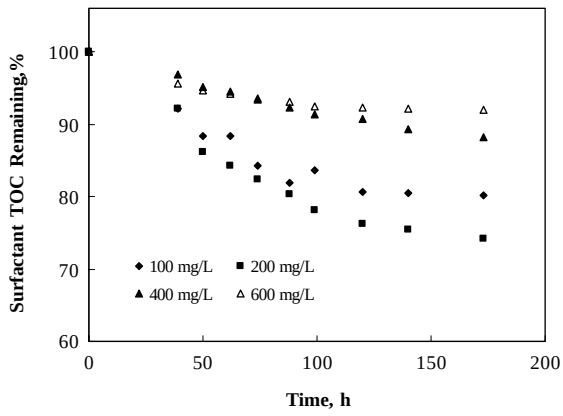

Figure 2. Biodegradation of Tergitol 15-S-7 at different surfactant concentrations (initial biomass – 0.22 mg carbon/l).

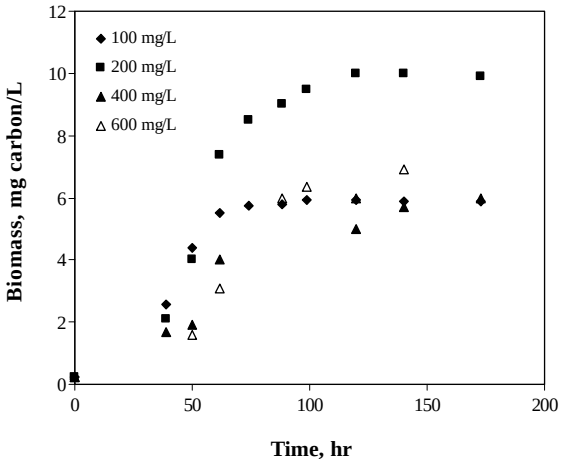

Figure 3. Effect of surfactant concentrations on bacterial growth, corresponding to the experimental results shown in Figure 2.

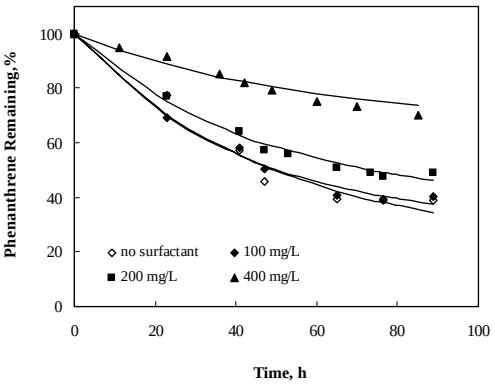

Figure 4. Effect of surfactant concentrations on biodegradation of phenanthrene at an initial phenanthrene concentration of 0.70 mg/l (initial biomass – 0.22 mg carbon/l). *Solid lines indicate the fitted results with Equation (2).

100, 200 and 400 mg/l, respectively. The initial concentration of phenanthrene in these experiments was 0.70 mg/l which equals to the aqueous solubility of phenanthrene. It appears that a surfactant concentration of 100 mg/l did not influence phenanthrene biodegradation. However, with further increases of the surfactant concentrations, the biodegradation rate and biodegradability of phenanthrene in the medium decreased. Similar phenomenon for biodegradation of phenanthrene was also reported for other surfactant/bacteria combinations in literature (Grimberg & Aitken 1995; Guha & Jeffe 1996).

The results in Figure 5 were obtained at a greater initial phenanthrene concentration of 2.13 mg/l. In this case, most phenanthrene in the medium would dissolve in the micellar phase of the surfactant. Again, the results in Figure 5 also clearly show that the increase of the surfactant concentrations led to decreased biodegradation rate and biodegradability of phenanthrene. At a surfactant concentration of 100 mg/l, almost 95% or 2.03 mg/l phenanthrene in the medium was biodegraded after 100 h. In contrast, at a surfactant concentration of 400 mg/l, only about 40% or 0.85 mg/l of phenanthrene in the medium was biodegraded after the same period of time. Nevertheless, the amount of phenanthrene biodegraded was greater than 0.70 mg/l, which provides evidence that some phenanthrene in the micellar phase was indeed available to the bacteria for biodegradation.

To obtain a better understanding of the effect of surfactant on phenanthrene biodegradation, the corresponding bacterial growth results are shown in Figure 6. Despite that the biodegradability of phenanthrene decreased monotonically with the increase of surfactant concentrations (see Figure 5), it is found from Figure 6 that the maximum bacterial growth occurred at 200 mg/l surfactant concentration, and further addition of surfactant inhibited the bacterial growth.

Three possible mechanisms may be responsible for the observed results in Figures 4 and 5. Mechanism 1: Bioavailability of phenanthrene in

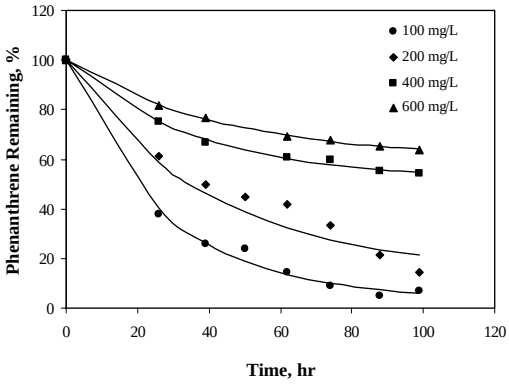

Figure 5. Effect of surfactant concentrations on biodegradation of phenanthrene at an initial phenanthrene concentration of 2.13 mg/l (initial biomass – 0.22 mg carbon/l). *Solid lines indicate the fitted results with Equation (2).

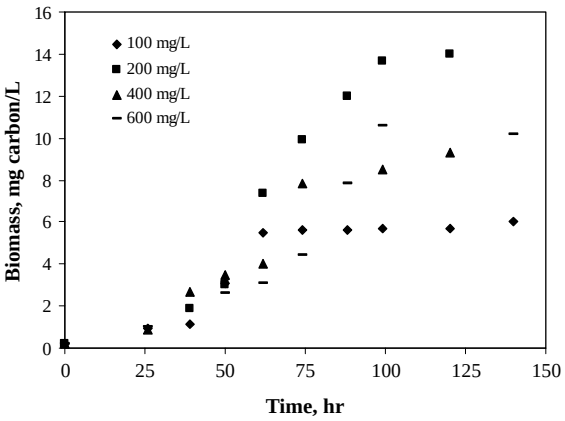

Figure 6. Bacterial growths during biodegradation of phenanthrene at different surfactant concentrations (initial phenanthrene concentration: 2.13 mg/l; initial biomass: 0.22 mg/l).

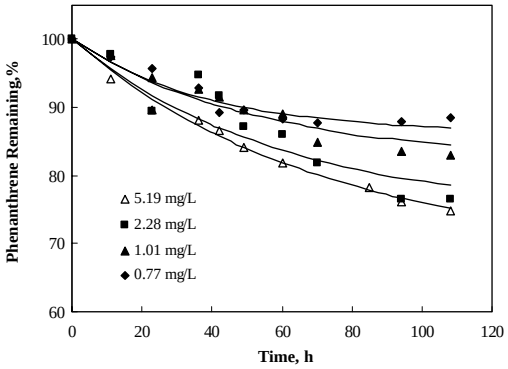

Figure 7. Biodegradation of phenanthrene at different initial phenanthrene concentrations (surfactant concentration – 550 mg/l, initial biomass – 0.22 mg carbon/l). * Solid lines indicate the fitted results with Equation (2).

the micellar solution reduced with the increase of the surfactant concentration. At higher surfactant concentrations, more phenanthrene molecules existed in the micellar phases, and the phenanthrene molecules were dispersed in a larger number of micelles and surrounded by more numbers or layers of surfactant molecules. In addition, higher surfactant concentrations may also result in the formation of more compacted micellar structures. All of these could cause the phenanthrene molecules in the micellar phase to be less biologically available at increased surfactant concentrations, due to the lower concentration gradient or mass transfer rate of phenanthrene from the micellar phase into the bacterial cells. Mechanism 2: Surfactant competed with phenanthrene as food source. Since the surfactant was biodegradable, the higher surfactant concentrations may cause the bacteria to rely much less on phenanthrene for their food. Therefore, both mechanisms 1 and 2 would suggest that biodegradation of phenanthrene can be adversely affected by the excess presence of the surfactant in the micellar solution. The third mechanism can be the inhibitory effect of high surfactant concentrations on bacterial activity, as implied in Figure 6.

Effect of initial phenanthrene concentrations on biodegradation

Figure 7 shows the results of biodegradation of phenanthrene at different initial concentrations in the medium with 550 mg/l of the surfactant. It is clearly demonstrated that, for the concentrations studied, the increase of the initial phenanthrene concentrations resulted in a greater biodegradability of phenanthrene. At a certain surfactant concentration, the increase of the initial phenanthrene concentration caused an increase of phenanthrene in the aqueous as well as in the micellar phases. The increase of phenanthrene in the aqueous phase was beneficial to biodegradation since phenanthrene in this phase was directly accessible to the bacteria, and hence had a greater bioavailability. Although the phenanthrene molecules in the micellar phase were generally less biologically available than those in the aqueous phase for biodegradation, the increase of phenanthrene concentrations in the micellar phase made the micellar phase more 'saturated' with phenanthrene. As a result, the phenanthrene molecules in the micellar phase were easier or had greater tendency to diffuse out into the aqueous phase when phenanthrene in the aqueous phase was consumed by the bacteria.

The results in Figures 4, 5, and 7 appear to suggest that phenanthrene biodegradation in the medium followed the first-order kinetics or an exponential decrease of the phenanthrene concentrations in the medium during the biodegradation process. If one assumes a first-order kinetics model as

$$\frac{dC}{dt} = -k(C - C^*) \quad (1)$$

Table 2. Parameter values obtained by fitting Equation (2) to the experimental data of phenanthrene biodegradation (initial biomass: 0.22 mg TOC/l)

Figure no.	Surfactant concentration (mg/l)	Initial phenanthrene concentration (mg/l)	k (h^{-1})	C^* (mg/l)
Figure 4	0	0.70	0.021	0.15
	100		0.025	0.20
	200		0.022	0.25
	400		0.019	0.45
Figure 5	100	2.13	0.037	0.07
	200		–[a]	0.31[b]
	400		0.028	1.14
	600		0.023	1.32
Figure 6	550	0.77	0.026	0.66
		1.01	0.019	0.83
		2.28	–[a]	1.75[b]
		5.19	0.014	3.53

[a] The data do not fit well to the first-order model.
[b] The data are taken or calculated from the final phenanthrene concentration.

the integration of Equation (1) leads to

$$C = C^* + (C_0 - C^*)e^{-kt} \quad (2)$$

where C_0 and C (mg/l) are the initial phenanthrene concentration and phenanthrene concentration at time t, respectively; C^* (mg/l) is the asymptotic phenanthrene concentration and k (h^{-1}) is the first-order rate constant.

Equation (2) is fitted to the experimental results in Figures 4, 5, and 7 and the fitted results are also shown in these figures in the solid lines. It is found that, in most cases, the biodegradation of phenanthrene in the experiments followed the first-order kinetics reasonably well. The values of k and C^* from the model fitting for the results are given in Table 2.

Effect of initial biomass on biodegradation of phenanthrene

Figure 8 shows the effect of the initial biomass on the biodegradation of phenanthrene at a surfactant concentration of 100 mg/l and an initial phenanthrene concentration of 1.05 mg/l. It is found that the increase of initial biomass concentrations from 0.24 to 0.96 mg carbon/l only slightly increased the biodegradation of phenanthrene.

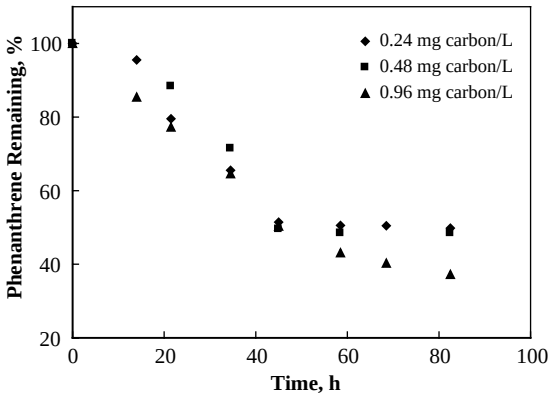

Figure 8. Effect of initial biomass concentrations on biodegradation of phenanthrene (surfactant concentration – 100 mg/l, initial phenanthrene concentration – 1.05 mg/l).

Biodegradability and bioavailability of phenanthrene

The experimental results presented above indicate the connection of biodegradation of phenanthrene (rate and extent) with its bioavailability in the micellar solution. The biodegradability of phenanthrene, B_d, may be expressed as:

$$B_d = 1 - \frac{C_f}{C_0} \quad (3)$$

where C_f is the final phenanthrene concentration in the medium at the end of the biodegradation process, and C_0 is the initial phenanthrene concentration in the medium. The biodegradability of phenanthrene versus the surfactant concentrations in the medium from the results obtained in Figures 4 and 5 are summarized in Figure 9. In general, the biodegradability of phenanthrene decreased almost linearly with the increase of the surfactant concentrations.

As aforementioned, the decrease in the biodegradability of phenanthrene with the increase of the surfactant concentrations can be, at least, partly attributed to the reduced bioavailability of phenanthrene in the medium. Since the solubility of phenanthrene in the micellar phase increases with the increase of surfactant concentrations in the medium, greater percentages of the total phenanthrene in the medium will be solubilized into the micellar phase. If one defines α as the solubilization extent of phenanthrene in the micellar phase, there are:

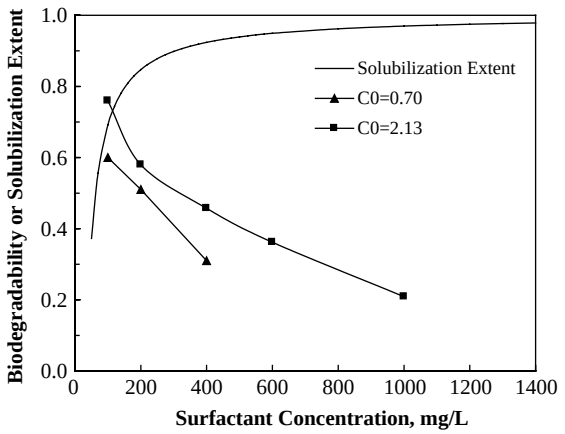

Figure 9. Biodegradability and solubilization extent of phenanthrene as a function of surfactant concentrations in the micellar solutions (initial biomass concentration – 0.22 mg carbon/l). * Lines with marks indicate biodegradability, and the line without markers indicates the solubilization extent.

$$\alpha = \frac{C_{\text{mic}}}{C} = 1 - \frac{C_{\text{aqu}}}{C} \tag{4}$$

$$C = C_{\text{mic}} + C_{\text{aqu}} \tag{5}$$

where C_{mic} (mg/l) is the concentration of phenanthrene in the micellar phase, C_{aqu} (mg/l) is the concentration of phenanthrene in the aqueous phase, and C (mg/l) is the apparent concentration of phenanthrene solubilized in the micellar solution (including both in the aqueous phase and in the micellar phase).

C_{mic} can be determined from the following equations as

$$C_{\text{mic}} = S_{\text{mic}} K_{\text{mw}} C_{\text{aqu}} \tag{6}$$

$$S_{\text{mic}} = S - \text{CMC} \tag{7}$$

where S and S_{mic} (mg/l) are the total surfactant concentration and the surfactant concentration that forms micelles in the micellar solution, respectively, and K_{mw} (l/mg) is the micelle–water partition coefficient of phenanthrene, defined as the ratio of the phenanthrene concentration in the micellar phase, C_{m} (equal to $C_{\text{mic}}/S_{\text{mic}}$), to that in the aqueous phase, C_{aqu}, (mg phenanthrene/l bulk solution), i.e.:

$$K_{\text{mw}} = \frac{C_{\text{m}}}{C_{\text{aqu}}} \tag{8}$$

From Equations (4)–(6), one can rewrite Equation (8) into:

$$K_{\text{mw}} = \frac{C_{\text{m}}}{C_{\text{aqu}}} = \frac{C_{\text{mic}}}{S_{\text{mic}} C_{\text{aqu}}} = \frac{\alpha C}{S_{\text{mic}}(1-\alpha)C} = \frac{\alpha}{S_{\text{mic}}(1-\alpha)} \tag{9}$$

The rearrangement of Equation (9) gives

$$\frac{1}{\alpha} = 1 + \frac{1}{K_{\text{mw}} S_{\text{mic}}} \tag{10}$$

Equation (10) indicates that the solubilization extent, α, increases with the surfactant concentration, S_{mic}, and is independent of the phenanthrene concentration, C. This is understandable since the distribution of phenanthrene between the two phases (i.e. the aqueous and micellar phases) is determined by the partition equilibrium and the amount of micelles in the micellar solution. The changes of α with the surfactant concentrations, as calculated from Equation (10), are also shown in Figure 9. It is demonstrated in this figure that the solubilization extent increases abruptly with the surfactant concentrations initially and then approaches the unity. This means that at a very high surfactant concentration, phenanthrene would reside mainly in the micellar phase, leaving only a negligible part in the aqueous phase. Since a substrate in the aqueous phase can be more readily bioavailable than the substrate in the micellar phase, the solubilization extent α may therefore be used as an indirect measure of the bioavailability of phenanthrene in the micellar solutions. Thus, for a constant phenanthrene concentration, the bioavailability of phenanthrene to biodegradation is potentially reduced with the increase of the surfactant concentrations, as observed in this study. Some other authors also reported a negative effect of higher surfactant concentrations on phenanthrene biodegradation (Grimberg & Aitken 1995; Guha & Jaffé 1996; Volkering et al. 1995).

Some final comments

The observations in this study are very meaningful to the application of surfactant in *in situ* bioremediation process. The advantage of using readily biodegradable surfactants is their low toxicity and nonaccumulation in the environment. However, their readily biodegradable nature also poses a disadvantage as microorganisms can use the surfactant as a possible carbon source. For poorly soluble polycyclic aromatic hydrocarbons

such as phenanthrene, the use of the surfactant Tergital 15-S-7 is found to largely enhance the solubility of phenanthrene in the micellar solution and make it possible for phenanthrene to be biodegraded by *Neptunomonas Naphthovorans*. At a constant initial phenanthrene concentration, the increase of the surfactant concentrations led to a decreased biodegradability of phenanthrene in the micellar solution. Within the solubility capacity of the surfactant, an increase of the initial phenanthrene concentration in the micellar solution enhanced the biodegradation of phenanthrene. The increase in biodegradability of phenanthrene can be attributed to the improved bioavailability of phenanthrene to microorganisms. In terms of bioremediation, while surfactant concentration should be high enough to achieve effective dissolution of the targeted compounds from a solid phase, such as soil, into a micellar solution for biological treatment, excess surfactant concentrations (more than that needed for dissolution) in the micellar solution should be minimized or avoided in order to obtain higher bioavailability and hence higher biodegradability of the compounds.

Acknowledgement

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