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Enhanced treatment of waste frying oil in an activated sludge system by addition of crude rhamnolipid solution

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ABSTRACT

The presence of high-strength oil and grease (O&G) in wastewater poses serious challenges for environment. Addition of surfactant into the activated sludge bioreactor is feasible in reducing high concentrations of O&G via enhancing its bioavailability. In this paper, an aqueous biosurfactant solution of rhamnolipid as a cell-free culture broth of *Pseudomonas aeruginosa* zju.um1 was added into a batch of aerobic activated sludge system for treatment of the waste frying oil. This treatment was conducted on both bench and pilot-scales, whereas the removal efficiency of frying oil was determined by analyzing the residue concentration of O&G and chemical oxygen demand (COD). In the presence of varying concentrations of rhamnolipid from 22.5 mg/L to 90 mg/L, aerobic treatment for 30 h was enough to remove over 93% of O&G while this biodegradability was only 10% in the control system with the absence of rhamnolipids. The equivalent biodegradability was similarly obtained on COD under addition of rhamnolipid. Compared with bench studies, a higher treatment efficiency with the presence of rhamnolipids was achieved on a pilot-scale of activated sludge system, in which a short time of 12 h was required for removing approximately 95% of O&G while the control treatment attained a low efficiency of 17%. Finally, foaming and biodegradability of rhamnolipids in activated sludge system were further examined in the whole treatment process. It seems that the addition of rhamnolipid-containing culture broth showed great potential for treatment of oily wastewater by activated sludge.

1. Introduction

It is reported that around 10 million tons of lipid-containing wastewater is discharged every year in China, most of which are from the food industries and kitchens. Oil and grease (O&G) in waste oil are usually clumped together, blocking the drain pipelines and grease traps so that they are not only easily removed but also resistant to the biotreatment of many microorganisms [1]. Hence, the presence of high-strength O&G in wastewater poses serious challenges for the environment. Biological treatment has been regarded as an efficient methodology to deal with the lipid-containing wastewater as well as other treatments including dissolved air flotation, flocculation and membrane filtration [2–6].

The addition of surfactants in aerobic or anaerobic activated sludge was regarded as a promising strategy for biotreatment of high-strength O&G. For example, Liu et al. found that the conventional aerobic sludge system could remove most of O&G at initial concentrations lower than 600 mg/L but, at higher concentrations of O&G, the addition of surfactant was required to facilitate the biodegradation [5]. The presence of surfactants was suggested to improve bioavailability of the hydrophobic substrate to the microbial via increasing their dispersion in aqueous environment or modifying the cell surfaces [7,8]. Surfactant was similarly effective in other biotreatment systems, including treatment of diesel oil by microorganisms in shaking flasks [9], aerobic treatment of dispersed diesel-containing wastewater in submerged biofilter [10], aerobic treatment of salad oil-containing wastewater in an immobilized continuous fluidized reactor [11] and anaerobic digestion of oil and grease-laden wastewater [12]. Being of low biodegradability, the chemically synthesized surfactant would accumulate in the industrial system as pollutants and, moreover, inhibit biodegradation of other carbohydrates via foaming and isolating air from sludge microbes. Thus, biosurfactants were more desirable for application in the environmental industries due to their specific capabilities of biodegradability and low toxicities.

However, the application of biosurfactants in industries is limited by its high cost compared with the chemically produced surfactants which were related to the costly raw material, low product

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concentration and complicated isolation. Commonly, biosurfactants are microbially produced [13]. Being one of such products, rhamnolipid has been well known for its high effectiveness and high expense as well, with the latter limiting its practical applications on a large scale [7,14,15]. Except for the economic concern, detrimental foam formation is another major factor rendering the limited use of biosurfactants in aerobic activated sludge treatment [16].

In our previous study, the waste frying oil was used as the sole carbon source to cultivate *Pseudomonas* sp. zju.um1 for producing rhamnolipid [17]. And the cost of the free-cell culture broth containing 35 g/L rhamnolipids was only 2800 USD/m^3. This paper addressed the feasibility of directly applying the rhamnolipid-containing culture broth for enhancing the biodegradation of waste frying oil in a conventional aerobic activated sludge system. Fermentation of waste oil for producing valuable products for wastewater treatment and the direct use of culture broth for saving the isolation cost would largely benefit the environment. To this goal, both bench and pilot studies were employed to evaluate the feasibilities regarding the removal efficiency as well as foaming and retention of rhamnolipids.

2. Materials and methods

2.1. Fermentation of rhamnolipids

Rhamnolipids were produced by *Pseudomonas* sp. zju.um1 which was previously isolated in our laboratory [17]. A 4% (m/v) concentration of waste frying oil was used as the sole carbon source. The composition of the culture medium was as follows (g/L); $NaNO_3$, 4.0; NaCl, 1.0; KCl, 1.0; $CaCl_2{\cdot}2H_2O$, 0.1; KH_2PO_4, 3.0; $Na_2HPO_4{\cdot}12H_2O$, 3.0; $MgSO_4$, 0.2; $FeSO_4{\cdot}7H_2O$, 0.001 and 2 mL of the trace element solution, which contains (per liter) 0.08 g $FeCl_3{\cdot}6H_2O$, 0.75 g $ZnSO_4{\cdot}7H_2O$, 0.075 g $CuSO_4{\cdot}5H_2O$, 0.75 g $MnSO_4{\cdot}H_2O$ and 0.15 g H_3BO_3. The final pH of each medium was set at 6.8. Rhamnolipids were fermented in a 50 L reactor with a working volume of 25 L, operating with an exterior foam recycling system. The culture was conducted at a stirring speed of 300 rpm and an aeration rate of 10 L air/min at 35–37 °C. At the end of fermentation (96 h), the culture broth was centrifuged at a speed of 5000 × *g* for 30 min and the supernatant cell-free culture broth was obtained. This clear culture broth was analyzed and shown to contain 35 g/L of rhamnolipids. The critical micelle concentration (CMC) of this cell-free culture broth was detected to be 28.8 mg rhamnolipids/L with a minimal surface tension of 30.1 mN/m by the ring method. Two components of rhamnolipids were identified as α-rhamnopyransyl-β-hydroxydecanoyl-β-hydroxydecanoate (Rha-Cl0Cl0) and 2-O-α- rhamnpyranosyl-β-hydroxydecanoyl-β-hydroxydecanoate (Rha-Rha-C10Cl0) [17].

2.2. Activated sludge and synthetic oily wastewater

The activated sludge was provided by the Ninbo DEAN Zoology Group. In each treatment, the mixed liquor suspended solid (MLSS) of the activated sludge was set at around 5400 mg/L with the sludge volume index (SVI) of around 25 g/L.

Waste frying oil was collected from the company cafeteria (Ningbo Dean Group Co, Ningbo, China) which had been used for frying sea fish and pork. The synthetic oily wastewater was prepared by mixing tap water and waste frying oil together at the final O&G of about 900 mg/L and COD of about 3600 mg/L.

2.3. Benchscale activated sludge system

The benchscale activated sludge treatment was conducted in glass tanks with a working volume of 4 L as shown in Fig. 1. Each glass tank was filled with 4 L of frying oil containing wastewater and aerated through two air distributors at a flow rate of 0.3 L air/min. The porosity of the sparger was 0.05–0.1 mm. Both distributors oppositely sat at the bottom of each tank. The temperature of the room was controlled at either 27 °C or 20 °C by using an air conditioner. The temperature of 27 °C was selected because it is the typical local room temperature while 20 °C was selected for representing the lower temperature. The volume of evaporated water was examined every 12 h and the additional tap water was correspondingly supplemented.

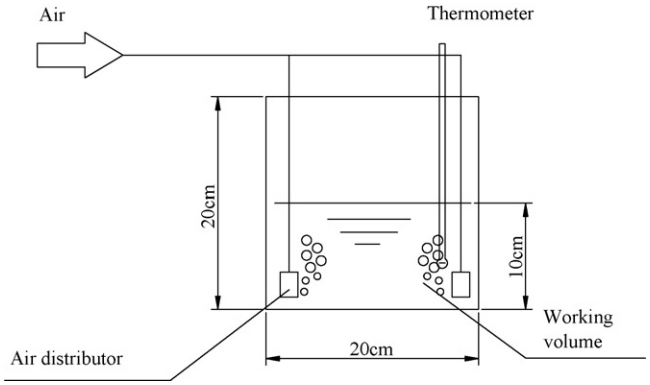

Fig. 1. Schematic diagram of aerobic activated sludge treatment system on bench scale.

In our preliminary experiment, the suitable rhamnolipid concentration for treating the wastewater has been roughly screened by using shaking flasks at a stirring speed of 120 rpm and found to be within the range of 20–100 mg/L (the data are not shown).

2.4. Pilot-scale activated sludge system

The batch aerated activated sludge system on pilot-scale was composed of a pool with a length of 2 m, a width of 0.5 m and a depth of 3 m, locating at the Ningbo DEAN Zoology Group (Ningbo, China). Two pools were arranged in parallel with each filled by 1 m^3 of oily wastewater with or without addition of rhamnolipid-containing culture broth. One Fine Bubble Diffusion Aerator sitting at the bottom provided the aeration and mixing inside the pool at a rate of 7.5 m^3 air/h. The operation was conducted at room temperature of 27–29 °C.

2.5. Analysis of residual O&G and COD

The samples of wastewater were specifically taken according to the emulsification extent in oily wastewater. Without addition of rhamnolipid-containing culture broth, oil and grease floated on wastewater making homogeneous sampling unavailable. Thus, in the bench studies without presence of rhamnolipids, all the final treated wastewater were taken as samples after stopping aeration for about 2 h when the activated sludge completely settled down. For each sampling in the control pilot-scale studies, a total of five samples with each volume of about 500 mL were collected at the four corners and the middle and were later combined them together. In presence of added rhamnolipid-containing culture broth, the activated sludge system was conveniently sampled due to a homogenous emulsification after aeration for 2 h. Thereafter, the wastewater with addition of culture broth was sampled at periodic intervals. The collected oily wastewater was put into the wide-mouth glass bottle with a teflon cap and acidified to pH lower than 2 by adding 2.5 ml of 10N H_2SO_4. The acidified samples were stored at 4 °C.

Table 1
Foam formation in activated sludge system.

Rhamnolipids concentration (mg/L)	0	22.5	45	90
Foam formation in activated sludge with rhamnolipids and waste frying oil	None	None	Less severe lasting for 20 min	Severe Lasting for 2 h
Initial surface tension in pure water (mN/m)	72.8 ± 2.8	31.2 ± 2.1	29.6 ± 4.8	29.0 ± 2.3
Initial surface tension in activated sludge system in absence of waste frying oil (mN/m)	71.8 ± 1.8	39.9 ± 2.2	36.1 ± 3.9	32.4 ± 1.8
Initial surface tension in activated sludge in presence of waste frying oil (mN/m)	60.5 ± 2.9	40.6 ± 3.3	37.9 ± 2.4	33.3 ± 2.1
Initial surface tension in formaldehyde-treated activated sludge in the absence of waste frying oil (mN/m)	54.0 ± 4.8	38.7 ± 2.8	35.3 ± 2.5	32.3 ± 3.5

Note: The initial surface tension in activated sludge system was measured after aeration for 30 min. Some data shown in Fig. 6 were also included in this table for convenient comparisons. The data are mean ± S.E.M. of 3 independent experiments.

Upon analysis, the samples were centrifuged at 1000 × *g* for 30 min to remove sludge. The O&G concentration and COD in oily wastewater were respectively estimated according to previous publications [18]. The O&G concentration was determined by extraction with tetrachloroethylene, and infrared detection of O&G at 220 nm. COD was analyzed using COD reagent vials, a COD reactor and a Lambda 20 UV–visible spectrophotometer. The removal efficiency was calculated as a relative percentage of decrease in either the initial O&G or COD.

2.6. Surface tension measurement

Surface tension was determined with a JYW-200A tensiometer (Chengde, China) using the ring method [19].

2.7. Measurement of rhamnolipids retention

The MLSS of the activated sludge was set at around 5400 mg/L while the SVI was around 25 g/L. Immediately after aeration for 30 min in all systems at varying rhamnolipid concentrations, the samples were collected every 6 h thereafter and centrifuged at 1000 × *g* for 30 min to get supernatant for detecting surface tension. In order to examine the possibility of biodegradability of rhamnolipids by sludge microbes, the activated sludge was pretreated by formaldehyde at a dose of 1.9% (v/v) for 30 min to inhibit microbial activities before the residual rhamnolipids contents were detected in the absence of waste frying oil.

The residual rhamnolipid contents were roughly estimated by detecting the surface tension of the aqueous solution in the benchscale activated sludge systems with or without the presence of waste oil (900 mg/L). As a calibration, analysis by HPLC, a Shimadzu HPLC LC-10AT with a UV detector SPD-10AV at 280 nm and a C18 column, was used to examine the correlation between aqueous rhamnolipids concentration and surface tension. The mobile phase comprised of acetonitrile and 10 mM ammonium acetate (60:40, v/v) and was run at 1 mL/min [19]. As expected, the surface tension was approximately linear with the aqueous rhamnolipid content at concentrations of rhamnolipids lower than CMC and, as a result, surface tension was higher than 35 mN/m.

2.8. Data analysis

All values for benchscale studies are mean ± stand error of mean (S.E.M.) of 3 independent experiments while the data for the pilotscale studies represent 2 independent experiments.

3. Results and discussion

3.1. Bench studies

The effect of rhamnolipids treatment on biodegradation of waste oil was first conducted on benchscale at 27 °C. Four such reactors were simultaneously run for comparisons with the initial rhamnolipid concentration ranging at 0, 22.5, 45 and 90 mg/L, respectively. The foaming upon aeration was recorded in Table 1. As indicated in Table 1, both the severity and the lasting period of foaming increased with the rhamnolipids dose. Immediately after aeration, a layer of white foam can only be observed in the bioreactors with the rhamnolipids dose of either 45 mg/L or 90 mg/L. However, the foam disappeared quickly after 20 min of running in the reactor with rhamnolipids of 45 mg/L and this time period of foaming was 2 h for the reactor at a rhamnolipid dose of 90 mg/L. In the control reactor without rhamnolipids, oils were observed to float on the top of aqueous solution in the whole treatment process while no obvious floating oil was noticed after 10 h in the reactor with rhamnolipids addition of 45 mg/L and 12 h for the other two reactors.

After aerobic treatment for 40 h, the samples in each reactor were collected for analysis of the final residual O&G concentration and COD. The results are shown in Fig. 2, which indicated that the high residual frying oil with O&G of 836 ± 96 mg/L and COD of 3481 ± 353 mg/L was detected in the control treatment without addition of rhamnolipids. Different from the control treatment, the residual O&G concentration reached 64.7 ± 8.6 mg/L, 44 ± 9.2 mg/L and 62 ± 7.6 mg/L for the treatment with rhamnolipids dosing at 22.5 mg/L, 45 mg/L to 90 mg/L, respectively. The final

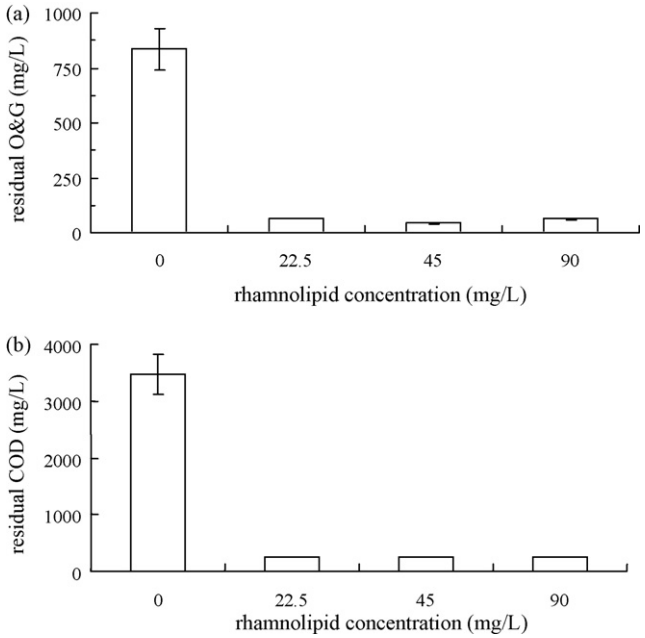

Fig. 2. The removal efficiency of waste frying oil in benchscale activated sludge system. The aerobic treatments were conducted in activated sludge system for 40 h under varying rhamnolipid doses varying from 0 to 90 mg/L. (a) the residual oil and grease concentration; (b) the residual COD. The data are mean ± S.E.M. of 3 independent experiments.

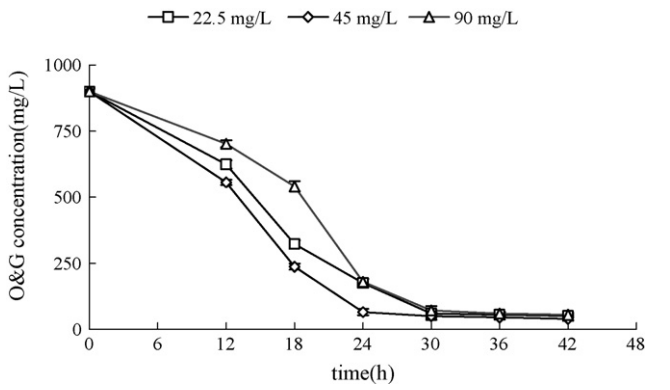

Fig. 3. The time course of residual oil and grease in benchscale activated sludge systems at varying rhamnolipid doses. The data are mean ± S.E.M. of 3 independent experiments.

COD in each biotreatment with rhamnolipids decreased to around 240 ± 1.6 mg/L. As the initial O&G concentration and COD were individually detected to be 912 mg/L and 3620 mg/L, the removing efficiency of O&G and COD was both calculated to be over 93% in the rhamnolipid groups and less than 10% in the control group. This result suggested that the addition of rhamnolipid-containing culture broth enhanced the emulsification of O&G and, as a result, greatly improved the removal efficiency.

The time course of biodegradation of O&G with addition of biosurfactant was then investigated under the same temperature of 27 °C as above. The comparable experiment with the control reactor was not included due to the difficult sampling with floating oil in the whole treatment. As the complete emulsification required 12 h of aeration for all three treatments with rhamnolipids, the wastewater in each bioreactor was sampled after treatment of 12 h. As shown in Fig. 3, a treatment time of 24 h was required for O&G removal efficiency reaching over 93% at a rhamnolipid dose of about 45 mg/L while for the other two bioreactors 30 h of treatment was needed to biodegrade most of the O&G. Hence, of the three treatments by rhamnolipids, biodegradation performed the fastest for a rhamnolipid dose of 45 mg/mL and lowest for rhamnolipids dose of 90 mg/L. The biodegradation occurred at the later stage of treatment for each bioreactor. With respect to the differential biodegradation rates among the three doses, we postulated that an addition of rhamnolipid (90 mg/L) might inhibit the biotreatment via reducing the mass transfer by foam formation within the first 2 h of treatment and the low dose of rhamnolipid (22.5 mg/L) might not provide a sufficient emulsification of the oil and grease [20]. It can also be concluded that the rate of biodegradation depends on the dispersion of O&G and is unrelated to biosurfactant dose.

The effect of rhamnolipid on biotreatment was examined again at a lower temperature of 20 °C. It was found that under this low temperature the O&G was less degradable compared with that at 27 °C and the residual O&G reluctantly reached to 100 ± 7.6 mg/L after four days of treatment in the presence of rhamnolipid (45 mg/L). But the situation in the control treatment without addition of rhamnolipid was even worse whereas the O&G floated on the surface within the first 3 days and later gradually turned into white solids. The increase in solubility of the O&G and the microbial's activity at high temperature could facilitate to interpret the enhanced biodegradation of waste oil at 27 °C. According to the previous findings the white solids were suggested as mixtures of calcium stearate and calcium palmitate, which was most possibly due to the saturated fatty acid released from triglycerides [11]. Interestingly, no such white solids were noticed in the comparable treatments with addition of rhamnolipids, corresponding well with the previous results that white solid was unnoticed with alkyl ether sulfate-based commercial surfactants added for treating the O&G [11]. It seems that the dispersion of saturated fatty acids by surfactants was regarded as the major factor for enhancing the removal efficiency. With dispersion by surfactants, most of O&G was readily broken down by microorganisms into carbon dioxide and water rendering it harmless to the ecosystem. This study did not examine the biodegradation under a high temperature around 35 °C in view of its low possibility on industrial scales because extra heating would not be used for maintenance of such temperature.

Fig. 4. Pilot treatment of waste frying oil. (a) the sampled slurry after aerated for 6 h, with the left from the activated sludge system without presence of rhamnolipids and the right from the system with presence of rhamnolipids (45 mg/L); (b) the photograph of the two activated sludge systems after the aeration was stopped for 2 h at the end of treatment. The left pool was without rhamnolipids, and the right pool was with 45 mg/L of rhamnolipids.

3.2. Pilot studies

The pilot studies were conducted in the activated sludge pools. The oily wastewater was composed of waste frying oil at a concentration of around 900 mg/L similar to the bench experiment. The culture broth with a volume of 1.28 L was added in the wastewater with rhamnolipid controlled around 45 mg/L. The room temperature varied between 27 and 29 °C.

Upon addition of the rhamnolipid-containing culture broth, a mass of foam was formed immediately floating on the surface of the sludge pool. Two hours later, the foam was gradually disappeared and no obvious floating O&G was noticed on the surface of aerated aqueous solution. In the control pool without addition of rhamnolipid-containing culture broth, the O&G initially floated as large patches on the surface and later appeared greasier. Fig. 4a

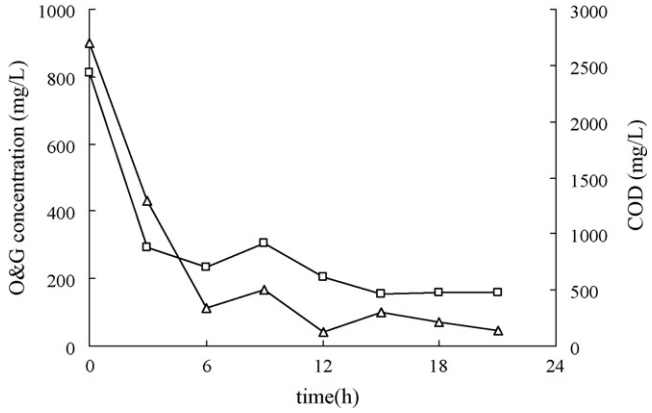

Fig. 5. The time course of residual oil and grease and COD in the pilot-scale activated sludge system. The data represents 2 independent experiments.

reflected the appearance of the activated sludge system at treatment for 6 h, whereas oil was heavily floated on top of the left flask without rhamnolipids and no accumulated oil drop was observed in the right pool with presence of rhamnolipids. At 22 h of treatment, the photo was taken after the aeration stopped and settled down for 1 h. As shown in Fig. 4b, an oily substance still floated on the surface of the control pool in the absence of rhamnolipids while no floating oil was evidenced on the right pool added with rhamnolipids, indicating the positive function of rhamnolipids in biotreatment of oily wastewater. By analysis of residual O&G and COD, the treatment with rhamnolipids reached the final removal efficiency of 95% for O&G and over 94% for COD while these values were only 17% and 21% for the control group.

The dynamic alterations of O&G and COD were presented in Fig. 5. As indicated in Fig. 5, the residual O&G concentration was under 42 mg/L after 12 h while no obvious biodegradation of the O&G was noticed without addition of rhamnolipids. In comparison to the bench studies, much faster degradation was obtained in this pilot study which could be due to much better aeration in this pilot system. The performance of COD degradation within 12 h of treatment was similarly presented in Fig. 5. The increase in COD after 12 h of treatment might be caused by the excretion of the microorganisms.

According to the dose of rhamnolipid at 45 mg/L, the addition of rhamnolipids was calculated to cost about 0.2 USD/m^3. As the common wastewater treatment by aerobic sludge system usually costs about 0.1–0.2 USD/m^3 in China, it was estimated that the estimated cost of applying rhamnolipid in treatment of the oily wastewater would be lower than 0.5 USD/m^3.

3.3. Foam formation of rhamnolipid in activated sludge

As mentioned in bench and pilot studies above, foam formation was not observed in activated sludge treatments added with rhamnolipid of 22.5 mg/L (close to the CMC of 28.8 mg/L) and was only noticed at the beginning for a short period for the other two doses, 45 mg/L and 90 mg/L, which are nearly in double and triple of the CMC, respectively. Interestingly, if rhamnolipid-containing culture broth at those amounts were added to pure water, severe foams would be noticed and maintained for a long time upon aeration. Also, biosurfactant solution at concentrations larger than the CMC was supposed to have high surface energy for foaming upon aeration. To address this contradictory phenomenon, surface tensions were immediately detected after a short aeration of 30 min in various activated systems with or without the presence of waste oil or formaldehyde-pretreated sludge. For comparison, rhamnolipid solutions of pure water were prepared by dissolving the same amounts of rhamnolipids into deionized water and their surface tensions were then detected upon aeration for 30 min. As shown in Table 1, with the same amount of rhamnolipids, the surface tensions of rhamnolipid solution of pure water were significantly lower than those in various activated sludge systems with or without the presence of waste frying oil. Despite the high initial doses higher than the CMC, the detected surface tensions were all above the critical surface tension at the CMC and, as a result, the rhamnolipid concentrations were estimated to be lower than the CMC. In this regard, we postulated that the adsorption of rhamnolipids by sludge or waste frying oil together with the biodegradation of rhamnolipids could explain the increase in surface tensions in activated sludge system.

However, in the absence of rhamnolipids, the initial surface tension in pure water was unexpectedly higher than those in the other sludge systems. At this point, we premised the excretion by sludge microbial community or some components in waste frying

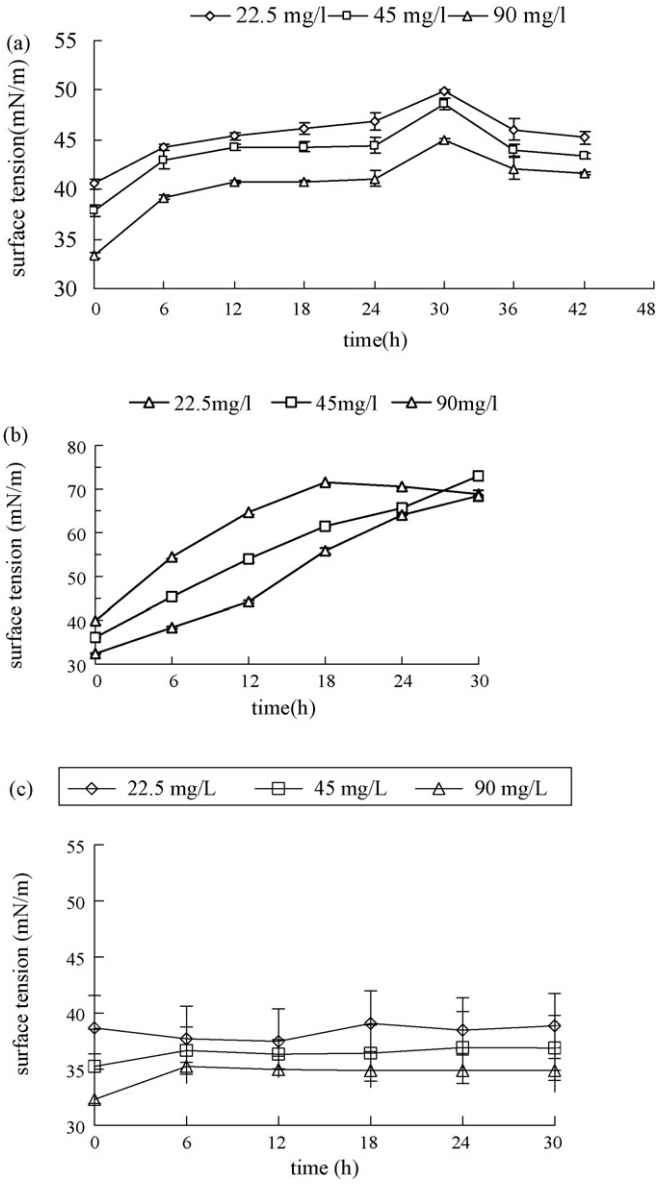

Fig. 6. The surface tensions of benchscale activated sludge systems. (a) the time course of surface tension in activated sludge in the presence of waste frying oil; (b) the time course of surface tension in activated sludge in the absence of waste frying oil; (c) the time course of surface tension in formaldehyde-treated sludge system in the absence of waste frying oil. The data are mean ± S.E.M. of 3 independent experiments.

oil might decrease the surface tension under the situation without mediation of extra rhamnolipids. The pretreatment by formaldehyde could be more effective in reducing the surface tension to a larger extent via promoting the excretion of intracellular biosurfactants. To be mentioned, this attribution on reducing surface tension by excretion of intracellular substance or waste frying oil would be a minor factor in comparison to the increase in surface tension by rhamnolipid adsorption.

3.4. Retention of rhamnolipids in activated sludge

Usually surfactant retention is not desirable due to environmental concerns and is thus regarded as an important factor in various applications. To estimate the biodegradability of rhamnolipid, its retention in aerobic activated sludge system was measured. For convenient analysis of rhamnolipids, the close relationship between the surface tension and biosurfantant concentration at a range lower than CMC could be roughly used to estimate the biosurfactant concentration as in previous work [21]. Hereby, the surface tension of wastewater was measured during the biodegradation in activated sludge at bench scale.

The retention of rhamnolipids was first examined in the benchscale activated sludge system with the presence of varying rhamnolipids additions while waste frying oil was fixed at an initial concentration of around 900 mg/L. As shown in Fig. 6a, the surface tension initially increased rapidly during the initial 6 h of treatment and then approached to a platform later. From this result, it could be seen that some rhamnolipids remained in aqueous solution while some were adsorbed to the sludge and waste oil.

The biosurfactant might be degraded in the absence of extra carbon source. Hence, the retention of rhamnolipids was also investigated without the presence of frying oil in simulation of the post-treatment of activated sludge system for removing surfactants after oily carbons were used up. Without the presence of waste frying oil, the surface tension, as expected, continuously increased to 65–70 mN/m which is close to the surface tension of pure water (72.8 mN/m). It was premised that without the presence of oil the added rhamnolipid was used as carbon source by microorganism, resulting in a drastic increase in surface tension. It seems that rhamnolipids could be biodegraded by the sludge microbial communities and thus of high environmental-friendliness. To further confirm the biodegradability of rhamnolipids by sludge microbes, the surface tensions were similarly measured in the formaldehyde-treated activated sludge systems. As indicated in Fig. 6c, pretreatment by formaldehyde sustained low surface tensions which demonstrated that inactivated microbes after formaldehyde could not take rhamnolipids as carbons source. This result supported the biodegradability of rhamnolipids by microbes.

Overall, it was shown that the removal of rhamnolipids from the aqueous phase of activated sludge caused the increase in surface tension in the activated sludge system. This decrease in rhamnolipids concentration in the aqueous phase could be due to the adsorption of activated sludge as well as waste oil [22] or the biological utilization as carbon sources by the microorganism [23]. This adsorption of rhamnolipids could largely cause the initially rapid decrease in surface tension in the beginning upon aeration while the microbial biodegradation of rhamnolipids dominated the decrease in surface tensions at a late stage when the oily carbon source was consumed.

4. Conclusions

Both the applications of waste frying oil as raw materials for fermentation of rhamnolipids and the further direct use of cell-free rhamnolipid-containing culture broth as biosurfactant solution largely cut down the cost of the final product of rhamnolipids and enhanced the feasibility of rhamnolipids in biotreatment of waste oil. Moreover, both bench and pilot studies illustrated that the addition of rhamnolipid-containing culture broth could greatly improve the efficiency of biodegradation of O&G in an aerobic activated sludge system. The foam formation and its subsequent deleterious effect on biodegradation of frying oil could be avoided by using low doses of rhamnolipids under 90 mg/L. Finally, the biodegradability of rhamnolipids was verified by detecting surface tensions. Both bench and pilot studies indicated great potential of rhamnolipids for treatment of wastewater by an aerated activated sludge system. Nevertheless, many other factors will have to be considered in application of rhamnolipids on a field-scale which include ionic strength of the oily wastewater and aeration rate.

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